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# PROCEEDINGS

# OF PAKISTAN CONGRESS OF ZOOLOGY

Volume 40 (2020-2021)



# FORTIETH PAKISTAN CONGRESS OF ZOOLOGY

MARCH 10-12, 2020

AND FIRST VIRTUAL PAKISTAN CONGRESS OF ZOOLOGY DECEMBER 14 AND 17-19, 2021

HELD UNDER THE AUSPICES OF THE ZOOLOGICAL SOCIETY OF PAKISTAN

AT

SINDH AGRICULTURE UNIVERSITY, TANDOJAM

PK ISSN 1013-346 CODEN: PKCZEK7

# **PROCEEDINGS**

### **OF**

# PAKISTAN CONGRESS OF ZOOLOGY

Volume 40, 2020

All research articles in this Proceedings were refereed by experts in respective disciplines



# FORTIETH PAKISTAN CONGRESS OF ZOOLOGY March 10-12, 2020 and First Virtual Pakistan Congress of Zoology December 14 and 17-19, 2021

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THE ZOOLOGICAL SOCIETY OF PAKISTAN

at

SINDH AGRICULTURE UNIVERSITY, TANDOJAM

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#### PROCEEDINGS OF THE CONGRESS

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Composed & Designed by: Fakhar Mahmood Shahid

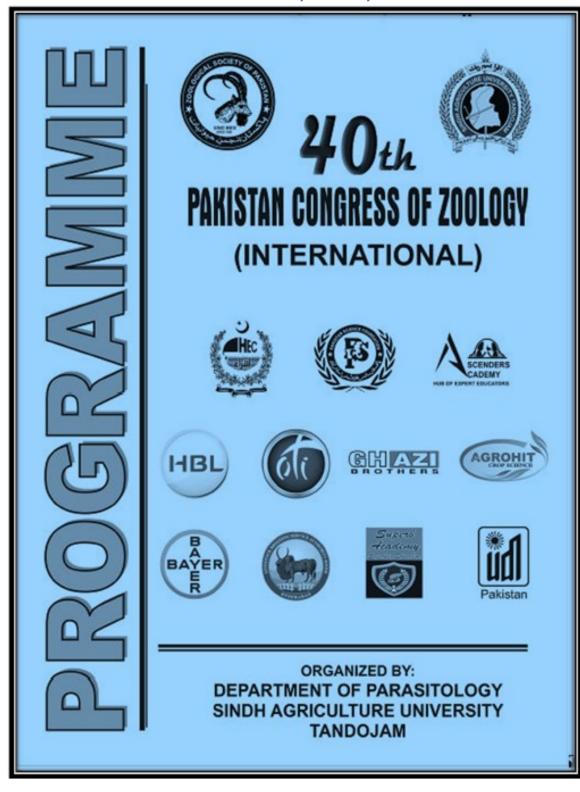
### **ACKNOWLEDGMENTS**

Department of Parasitology, Sindh Agriculture University, Tandojam hosted the 40<sup>th</sup> Pakistan Congress of Zoology (International).

The Zoological Society of Pakistan expresses its deep gratitude to the Vice Chancellor, Sindh Agriculture University, Tandojam, faculty members and students of the Department of Parasitology for extending warm hospitality.

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# First Virtual Pakistan Congress of Zoology December 14, 17-19, 2021



# First Virtual Pakistan Congress of Zoology December 14, 17-19, 2021



# Inaugural Session (Hybrid Session) December 14, 2021 9:00AM to 5:00 PM



#### Dr. A.M. Shaikh Auditorium, Sindh Agriculture University, Tandojam

10:00 A.M. : Arrival of Presiding Panel

10:05 A.M. : Guests to be seated

10:10 A.M. : Recitation from Holy Quran.

10:20 A.M. : Welcome Address by Prof. Dr. Abdullah G. Arijo,

Chairman, Department of Parasitology, Sindh Agri. Univ, Tandojam

10:30 A.M. : Address of Mr. Abdul Aziz Khan,

Secretary General, Zoological Society of Pakistan

10:40 A.M. : Address of Prof. Dr. A.R. Shakoori

President, Zoological Society of Pakistan

10:50 A.M. : Address by Prof. Dr. Fateh Muhammad Marri

Vice Chancellor, Sindh Agriculture University, Tandojam.

11:00 A.M. : A Few Words by the Guest of Honor

11:05 A.M. : Awards distribution

11:30 A.M. : Address by the Chief Guest

11:45 A.M. : Vote of thanks by the Congress Secretary

11:50 A.M. : Announcements

11:55 A.M. : Refreshments

### 12:30 PM JOINT SESSION I: PLENARY LECTURES

Chairperson: Dr. Abdullah G. Arijo Co-Chairperson: Dr. Naeem Tariq Narejo

1. **Prof. Dr. Yan Zhang**, Key Laboratory of Aquatic Genomics, Ministry of Agriculture, and Beijing Key Laboratory of Fishery Biotechnology, Chinese Academy of Fishery Sciences, Beijing, 100141, China.

"Functional Characterization of Fatty Acid Desaturases (Fads2) In Common Carp (Cyprinus Carpio): Insights in to Catalyzing Efficiency Based On In Vitro, In Vivo And Docking Simulations"



**2. Prof. Dr. Telat Yanik,** Aquaculture Department, Fisheries Faculty, Ataurk University, 25240 – Erzurum, Turkey.

"Some Innovative Technologies in Aquaculture with Respect to Fish Feeds and Cage Farming"



3. **Prof. Dr. A.R. Shakoori,** School of Biological Sciences, University of the Punjab, Lahore. "Epithelial – Mesenchymal Transition: Effect of Metformin Chloride"



# 2:30 PM JOINT SESSION II: PLENARY LECTURES

Chairperson: Prof. Dr. Shahid Mahmood Baig Co-Chairperson: Dr. Riffat Sultana

 Prof. Dr. Abdullah G. Arijo, Department of Parasitology, Sindh Agriculture University, Tandojam.
 "Tick Control: Conventional Stratigies and Novel Approaches"



**2. Dr. Muge Hekimoglu**, Aquaculture Department, Ege University, 35100 Bornova, Izmir, Turkey. "The Effect of Extending Light Period in Fish Culture"



3. **Dr. Riffat Sultana,** Tea Research Institute of Chinese Academy of Agricultural Sciences (CAAS), Hangzhou 310008, China. "Historical Swarming of Locust in Pakistan: Causes and Control Strategies"





# Academic Sessions (Virtual Sessions) December 17-19, 2021 9: 00AM to 5:00 PM



# **DAY ONE**

# Friday, December 17, 2021

### 9:00 AM JOINT SESSION III: PLENARY LECTURES

Chairperson: Prof. Dr. A.R. Shakoori, Co-Chairperson: Prof. Dr. Javed Iqbal Qazi

 Prof. Dr. Aleksei Potekhin, Department of Microbiology, Faculty of Biology, Saint Petersburg State University, Saint Petersbrug, Russia. "Diversity, Speciation Mechanisms and the Species Concept In Paramecium (Ciliophora)"



2. **Prof. Dr. Muhammad Fiaz Qamar**, Department of Pathbiology, University of Veterinary and Animal Sciences, Jhang Campus, Jhang. "Prevalence of Eimeria and Capillaria Species in Field Studies of Pigeon (Columba Livia)"



3. Dr. Abdul Rehman, Institute of Microbiology and Molecular Genetics, University of the Punjab, Lahore.
"Microbacterium Sp. Strain 1s1 Resistance



"Microbacterium Sp. Strain 1s1 Resistance Strategies Against Metals: A Feasible Approach For Environmental Clean-Up"

# 11:00 - 1:00 PM

Chairperson: Prof. Dr. Farah R. Shakoori Co-Chairperson: Dr. Dilara A. Bukhari CELL BIOLOGY, MOLECULAR BIOLOGY, PHYSIOLOGY, BIOCHEMISTRY, GENETICS AND TOXICOLOGY		Chairperson: Prof. Dr. Naeem Tariq Narejo Co-Chairperson: Dr. Noor Khan FISHERIES, ECOLOGY, WILDLIFE, FRESHWATER BIOLOGY, MARINE BIOLOGY		
CBGP		FEWFM	-	
Farzana Iftikhar (WU, Multan)	CBGP-1	Wajiha Annum (LCWU, Lahore)	FEWFM-1	
Zainab Akram (PU, Lahore)	CBGP-2	Hira Arshad (LCWU, Lahore)	FEWFM-2	
Asma Waheed (GCU, Sialkot)	CBGP-3	Jemi (US, Jamshoro)	FEWFM-3	
Muhammad Afaq (MNSUA, Multan)	CBGP-4	Qandeel Minahal (LCWU, Lahore)	FEWFM-4	
Muhammad Tariq Zahid (GCU, Lahore)	CBGP-5	Wajeeha Komal (LCWU, Lahore)	FEWFM-5	
Naaz Abbas (PCSIR, Lahore)	CBGP-6	Wajeeha Komal (LCWU, Lahore)	FEWFM-6	
Chaman Ara, (PU, Lahore)	CBGP-7	Razia Liaqat (LCWU, Lahore)	FEWFM-7	
Khadium Mustafa (UA, Peshawar)	CBGP-8	Fatima Yasin (UE, Lahore)	FEWFM-8	
Muhammad Faraz Uddin (UK, Karachi)	CBGP-9	M. Mudassar Shahzad (UE, Lahore)	FEWFM-9	
Ayesha Sania (PU, Lahore)	CBGP-10	Faiza Shahid Wazir (UP, Peshawar)	FEWFM-10	
Asma Hassan (PU, Lahore)	CBGP-11	Amina Tahir (UVAS, Lahore)	FEWFM-11	
Tooba Zahid (PU, Lahore)	CBGP-12	Huma Anwar (UVAS, Lahore)	FEWFM-12	
Tuba Arooj (PU, Lahore)	CBGP-13	Aasma Noureen (VU, Pakistan)	FEWFM-13	
Ambreen Akram (FUUAST, Karachi)	CBGP-14	Dharti S. Thebo (US, Jamshoro)	FEWFM-14	
1:00 - 2:00 PM PRAYER BREAK (ZUHAR)				

Chairperson: Prof. Dr. Nasreen Memon Co-Chairperson: Dr. Abida Butt			
PESTS AND PEST CONTROL, ENTOMOLOGY AND PARASITOLOGY PC, ENT AND PAR			
Khizar Hayat (MNSUA, Multan)	PC-1		
Nosheen Jehajo (US, Jamshoro)	PC-2		
Sajid Siyal (US, Jamshoro)	PC-3		
Zaryab Gul (US, Jamshoro)	PC-4		
Saima Pathan (US, Jamshoro)	PC-5		
Aiman Amur (US, Jamshoro)	PC-6		
Maleeha Jamil (US, Jamshoro)	PC-7		
Noor Ul Huda Fatima (IU, Bahawalpur)	PC-8		
Aneela Jwaid Hssain (US, Jamshoro)	PC-9		
Raazia Tasadduq (KCW, Lahore)	PC-10		
Aiman Khalid (MNSUA, Multan)	PC-11		
Esha Mehik Fatima (MNSUA, Multan)	PC-12		
Saba haider (MNSUA, Multan)	PC-13		
Hina Ali (MNSUA, Multan)	PC-14		
1:00 – 2:00 PM PRAYER BREAK (Z	UHAR)		

# 2:00 - 4:45 PM

Chairperson: Dr. Abdul Rehman Co-Chairperson: Dr. Muhammad Khan		Chairperson: Dr. Noor us Sahe Co-Chairperson: Dr. Fa	rah Naz	
CELL BIOLOGY, MOLECULAR BIOLOGY, PHYSIOLOGY, BIOCHEMISTRY, GENETICS AND TOXICOLOGY CBGP		FISHERIES, ECOLOGY, WILDLIFE, F BIOLOGY, MARINE BIOLO FEWFM		
Sehreem Gill (KCU, Lahore) Samra Hasnat (PU, Lahore) Ayesha Liaqat (PU, Lahore) Ayesha Zafar (PU, Lahore) Hina Mahreen (WU, Multan) Afifa Yaqoob (PU, Lahore) Mehroze Amin (PU, Lahore) Jabbar Khan (GU, D.I. Khan) Sadia Arshid (UVAS, Lahore)	CBGP-15 CBGP-16 CBGP-17 CBGP-18 CBGP-19 CBGP-20 CBGP-21 CBGP-22 CBGP-22	Sadaf Khadim (UVAS, Lahore) Noor Hawa (UK, Karachi) Bibi Ayesha (UK, Karachi) Hina Jabeen (UK, Karachi) Muhammad Tabish (UK, Karachi) Arooba Nasir (UK, Karachi) Noor Us Saher (UK, Karachi) Dur e Shehwar (UK, Karachi) Farah Naz (UK, Karachi)	FEWFM-15 FEWFM-16 FEWFM-17 FEWFM-18 FEWFM-19 FEWFM-20 FEWFM-21 FEWFM-22 FEWFM-22	
Umair Ahmad (UVAS, Lahore) Sadia Mushtaq (PU, Lahore) Iqra Shahid (PU, Lahore) Naureen Fatima (GCU, Lahore) Shamsa Manzoor, (WU, Bagh, AJK)	CBGP-24 CBGP-25 CBGP-26 CBGP-27 CBGP-28	Wafra Matanat Zaheen (UK, Karachi) Syeda Hadiqa Noor (UK, Karachi) Altaf Hussain Narejo (UK, Karachi) Abdul Hameed (UK, Karachi) Roomana Yasmeen (UK, Karachi)	FEWFM-24 FEWFM-25 FEWFM-26 FEWFM-27 FEWFM-28	
4:45 – 5:00 PM PRAYER BREAK (MAGHRIB)				

Chairperson: Prof. Dr. Amjad Farooq Co-Chairperson: Dr. Nadir Ali Birmani			
PESTS AND PEST CONTROL, ENTOMOLOGY AND PARASITOLOGY PC, ENT AND PAR			
Kainat Iqra (MNSUA, Multan)	PC-15		
Mohsin Ali Jutt (US, Jamshoro)	PC-16		
Umer Sharif (MNSUA, Multan)	PC-17		
M. Salman Shafi (MNSUA, Multan)	PC-18		
Marwa (SBBWU, Peshawar)	PC-19		
Abro Zain-ul-Aabdin (US, Jamshoro)	PC-20		
Fatima Anum (US, Jamshoro)	PC-21		
Farheen Shaikh (US, Jamshoro)	ENT-1		
Kamal Khan Abro (US, Jamshoro)	ENT-2		
Sidra Tul Muntha (US, Jamshoro)	ENT-3		
Asif Nazeer Memon (US, Jamshoro)	ENT-4		
Ali Raza Soomro (US, Jamshoro)	ENT-5		
Asif Raza Soomro (US, Jamshoro)	ENT-6		
Asma Abdul Latif (LCWU, Lahore)	ENT-7		
4:45 – 5:00 PM PRAYER BREAK (MAGHRIB)			

### **DAY TWO**

# Saturday, December 18, 2021

### 9:00 - 01:00 PM

Chairperson: Prof. Dr. Syed Shahid Co-Chairperson: Dr. Iram	Chairperson: Prof. Dr. Muhammad Akhtar Co-Chairperson: Dr. Muhammad Akbar Khan		
CELL BIOLOGY, MOLECULAR BIOLOGY, PHYSIOLOGY, BIOCHEMISTRY, GENETICS AND TOXICOLOGY CBGP		FISHERIES, ECOLOGY, WILDLIFE, FF BIOLOGY, MARINE BIOLOG FEWFM	RESHWATER
Hooria Younas (KCW, Lahore)	CBGP-29	Venus A. Qadir (UK, Karachi)	FEWFM-29
Komal Fatima (KCW, Lahore)	CBGP-30	Tayyaba Hamid (UK, Karachi)	FEWFM-30
Khadium Mustafa, (UA, Peshawar)	CBGP-31	Kiran Aftab, (UG, Gujrat)	FEWFM-31
Ghulam Mustafa (GCU, Lahore)	CBGP-32	Muhammad Asim (PU, Lahore)	FEWFM-32
Sameena Gul (PU, Lahore)	CBGP-33	Muhammad Akbar Khan (PU, Lahore)	FEWFM-33
Amara Maryam (PU, Lahore)	CBGP-34	Sayyed Ghyour Abbas (PU, Lahore)	FEWFM-34
Aisha Ashfaq (SBBWU, Peshawar)	CBGP-35	M. Adeeb Babar (PU, Lahore)	FEWFM-35
Uzma Jabeen (FUUAST, Karachi)	CBGP-36	Z.B. Mirza (IUCN, Islamabad)	FEWFM-36
Qindeel Fatima (PU, Lahore)	CBGP-37	Fozia Afzal (IU, Bahawalpur)	FEWFM-37
Maryam Javed (UVAS, Lahore)	CBGP-38	Muhammad Khalil (UO, Okara)	FEWFM-38
Nadeem Raza (UVAS, Lahore)	CBGP-39	Muhammad Furqan (PU, Lahore)	FEWFM-39
Roheela Yasmeen (LGC, Lahore)	CBGP-40	Iqra Raees Shaikh (US, Jamshoro)	FEWFM-40
Bakhtawar Banori (SBBWU, Peshawar)	CBGP-41	Hira Lakho (US, Jamshoro)	FEWFM-41
Muhammad Waris (UVAS, Lahore) (For Poster) CBGP-42			

### 1:00 – 2:00 PM PRAYER BREAK (ZUHAR)

#### Chairperson: Prof. Dr. Shafqat Saeed Co-Chairperson: Dr. Mudassar Ali PESTS AND PEST CONTROL, ENTOMOLOGY AND **PARASITOLOGY** PC, ENT AND PAR Naheed Baloch (US, Jamshoro) ENT-8 M. Awais Ahmad (MNSUA, Multan) (For Poster) ENT-9 Danyal Haider Khan (MNSUA, Multan) **ENT-10** Shahid Iqbal (MNSUA, Multan) ENT-11 Kamran Ejaz (MNSUA, Multan) **ENT-12** Mudssar Ali (MNSUA, Multan) **ENT-13** Naila Bhanger (US, Jamshoro) ENT-14 Kavita Bai (US, Jamshoro) ENT-15 Muhammad Shafiq (MNSUA, Multan) **ENT-16** Alina Sabir (MNSUA, Multan) ENT-17 Sadique Ibrahim (MNSUA, Multan) **ENT-18** Muhammad Zubair (MNSUA, Multan) **ENT-19** M. Khalid Rafique (NARC, Islamabad) ENT-20 M. Talha Ahmad (MNSUA, Multan) ENT-21 1:00 - 2:00 PM PRAYER BREAK (ZUHAR)

#### 2:00 - 4:40 PM

Chairperson: Prof. Dr. Bushra Muneer Co-Chairperson: Dr. M. Tariq Zahid	Chairperson: Dr. Abdul Aleem Ch. Co-Chairperson: Dr. Tanveer Hussain			
CELL BIOLOGY, MOLECULAR BIOLOGY, PHYSIOLOGY, BIOCHEMISTRY, GENETICS AND TOXICOLOGY CBGP  Sobia Rizwana (PU, Lahore) CBGP-43  Nazia Haider (WU, Multan) CBGP-44  Summera Fatima (WU, Multan) CBGP-45  Muhtaq H. Lashari (IU, Bahawalpur) CBGP-46  Humera Afridi (GFCW, Peshawar) CBGP-47  Shama Sadaf (LCWU, Lahore) CBGP-48  N.Z. Ghumman (UVAS, Lahore) CBGP-49  Fouzia Tabassum (UE, Lahore) CBGP-50  Mudasar Hussain (UVAS, Lahore) CBGP-51  Ammara Younas (GCU, Lahore) CBGP-52	FISHERIES, ECOLOGY, WILDLIFE, FRESHWATER BIOLOGY, MARINE BIOLOGY FEWFM  Fakhra Nazir (CUST, Islamabad) FEWFM-42 Ali Umar (UO, Okara) FEWFM-43 Sangam Khalil (IU, Bahawalpur) FEWFM-44 Ali Hassan Rana (IU, Bahawalpur) FEWFM-45 Rubab Athar (IU, Bahawalpur) FEWFM-46 Muhammad Rais (AAU, Rawalpindi) FEWFM-47 Abu ul Hassan Faiz (WU, Bagh, AJK) FEWFM-48 Syeda Maria Bashir (WU, Bagh, AJK) FEWFM-49 Sarwan Khan (US, Jamshoro) FEWFM-50 Abdul R. Shaikh (US, Jamshoro) FEWFM-51 Namra Qaiser (UVAS, Lahore) FEWFM-52			
Shifa Rabbani (PU, Lahore) CBGP-53 Iram Liaqat (GCU, Lahore) CBGP-54 M. Zahid Farooq (CVAS, Jhang) CBGP-55 Nazneen Alam, (KIU, Gilgit) CBGP-56	M. Saim Ibtesam (MNSUA, Multa FEWFM-53 Shaista Jalbani UVAS Sakrand FEWFM—06-2020 Musarrat-ul-Ain (UK Karachi) FEWFM—15-2020 Munawwer Rasheed (CEMB Karachi) FEWFM—40-2020 Muhammad Ramzan (PU Lahore) FEWFM—61-2020			
4:45 – 5:00 PM PRAYER BREAK (MAGHRIB)				

# Chairperson: Prof. Dr. Abdul Mannan Sheikh Co-Chairperson: Dr. Santosh Kumar PESTS AND PEST CONTROL, ENTOMOLOGY AND

**PARASITOLOGY** PC, ENT AND PAR M. Younus (CUVAS, Bahawalpur) ENT-22 Bahadur Ali Gadani (SALU, Khairpur) **ENT-23** Summaya Naimat Shaikh (SALU, Khairpur) ENT-24 G. Mujtaba Khaskheli (SALU, Khairpur) ENT-25 Waheed Ali Panhwar (SALU, Khairpur) ENT-26 Ghulam Abbas Soomro (SALU, Khairpur) ENT-27 S.H. Masood Bokhari (MNSUA, Multan) ENT-28 Shabana Mangi (SALU, Khairpur) ENT-29 Mujeeb A. Memon SAU Tandojam PC-61-2020 ENT-7-2020 Farheen Deeba Soomro (US, Jamshoro) ENT-14-2020 Raheela Shah (US, Jamshoro) Saiqa Sanam (US, Jamshoro) ENT-16-2020 Gulalai Jamal Yousafzai (UMD KPK) PAR-26-2020 Noorul Hadi (HU Mansehra) PAR-27-2020 Tawseef Khan (VU Pakistan) PAR-28-2020

# **JOINT SESSION IV: PLENARY LECTURES**

Chairperson: Prof. Dr. A.R. Shakoori Co-Chairperson: Abdul Aziz Khan

**Dr. Muhammad Munir,** Biomedical and Life Sciences, Lancaster University United Kingdom.

"Zoonotic and Zooanthroponotic potential of Sars-Cov-2."



4:45 PM

#### DAY THREE

## Sunday, December 19, 2021

#### 9:00 - 11:00 AM

# Chairperson: Prof. Dr. Soumble Zulfiqar Co-Chairperson: Dr. Raazia Tasadduq

CELL BIOLOGY, MOLECULAR BIOLOGY, PHYSIOLOGY, BIOCHEMISTRY, GENETICS AND TOXICOLOGY

	CBGP-57
Ayesha Aihetasham, (PU, Lahore)	CBGP-58
Mehnaz Akhtar (GCW, Chakwal)	CBGP-59
Sana Nasim (WU, Bagh, AJK)	CBGP-60
Faiza Hakeem (WU, Bagh, AJK)	CBGP-61
Aisha Shah (US, Jamshoro)	CBGP-62
G.M.A. Ghani (Dera Ghazi Khan)	
Amna Mushtaq (WU, Multan)	CBGP-63
Adnan Ahmad (UA, Peshawar)	CBGP-64
Farhukh Tabasum (UA, Peshawar)	CBGP-65
	CBGP-66
Ayesha Hidayat (SBBWU, Peshawar)	CBGP-67
Asma (SBBWU, Peshawar)	CBGP-68
Zubaida Khanam Abbasi (Larkana)	CBGP-69
Imalah Habiba (UG, Gujrat)	CBGP-70
Rabia Sadiq (WU, Multan)	

### 1:00 - 2:00 PM PRAYER BREAK (ZUHAR)

#### Chairperson: Dr. Saima Naz Co-Chairperson: Dr. Waheed Ali Panhwar PESTS AND PEST CONTROL, ENTOMOLOGY AND **PARASITOLOGY** PC, ENT AND PAR Maree Rajper (US, Jamshoro) PAR-1 Ramesh (US, Jamshoro) PAR-2 Naheed Bano (US, Jamshoro) PAR-3 Bushra Khalil (Dera Ghai Khan) PAR-4 M. Zameer Kayani (GBP, Rawalpindi) PAR-5 Ali Asghar (AAU, Rawalpindi) PAR-6 Tariq Mukhtar (AAU, Rawalpindi) PAR-7 Sanam Samo (US, Jamshoro) PAR-8 Rabia Batool (GCU, Lahore) PAR-9 Raheela Noor Memon (US, Jamshoro) PAR-10 1:00 - 2:00 PM PRAYER BREAK (ZUHAR)

# 2:00 - 4:40 PM

# Chairperson: Dr. Muhammad Munir Co-Chairperson: Dr. Abdullah Shakir

### CELL BIOLOGY, MOLECULAR BIOLOGY, PHYSIOLOGY, BIOCHEMISTRY, GENETICS AND TOXICOLOGY

		CBGP-71
	aleeha Khalid (PU, Lahore)	CBGP-72
Ur	rooj Nazir (UH, KPK)	CBGP-73
Sa	ania Qaisar (UVAS, Lahore)	CBGP-74
Sa	arwar Allah Ditta (GCU, Lahore)	CBGP-75
Ra	aheema Tahir (UK, Karachi)	CBGP-76
Sh	nah Banish (SBBWU, Peshawar)	CBGP-77
Ra	abia Ejaz (PU, Lahore)	
Er	rum Zafar (PU, Lahore)	CBGP-78
Nι	udrat (SBBWU, Peshawar)	CBGP-79
	ohammad Attaullah (UM, Dir Lower)	CBGP-80
	. lazaz Kha (US, Swabi)	CBGP-81
		CBGP-82
	wais Niaz (US, Swabi)	CBGP-83
	abina Sardar (US, Swabi)	CBGP-84
Sa	addam Hussain (US, Swabi)	CBGP-85
W	/aqas (US, Swabi)	CBGP-86
Zu	unaiara Safdar (US, Swabi)	CBGP-4-2020
Sh	nafaat Yar Khan (US Sargodha)	CBGP-182-2020
Sa	aira Bano (UK Karachi)	050. 102 2020

# 4:45 - 5:00 PM PRAYER BREAK (MAGHRIB)

# **POSTERS SECTION**

Muhammad Ali (PU, Lahore)
Fakhr Ul Islam (US, Swat)
Hamza Umar (US, Swat)
POSTER-2
Huda Bilal (MNSUA, Multan)
POSTER-3
Bilal Kabeer (CULSP, Czech Republic)
Sadia Abid (CULSP, Czech Republic)
Arifa Savanur (UK, Karachi)

Faiza Khan (UK, Karachi)

POSTER-7 (POSTER-76-2020)

POSTER-8

Humaira Noor (UK, Karachi)

Tayyaba Hamid (UK, Karachi)

POSTER-9

Muhammad Waris (UVAS, Lahore)

POSTER-10 (POSTER-83-2020)

Wali Khan (UM, Malakand KPK)

POSTER-11(CBGP-42)

M. Awais Ahmad (MNSUA, Multan) (For Poster)

POSTER-12

Sidra Abbas (GDEC, Lahore)

POSTER-13 (ENT-13)

POSTER-14

# **CONCLUDING CEREMONY**

02:00 P.M. : Recitation from Holy Quran.

02:05 P.M. : Congress Report and Concluding Remarks by

Prof. Dr. A.R. Shakoori

President, Zoological Society of Pakistan

02:30 P.M. : Vote of thanks by the Congress Secretary

Prof. Dr. Abdullah G. Arijo,

Chairman, Department of Parasitology, Sindh Agri. Univ, Tandojam

#### MEMBERS OF THE CONGRESS

#### **BAHAWALPUR**

Batool, F.
Danish, D.
Habib, H.
Hameed, S.
Khalil, S. (Dr.)
Kumar, S (Dr.)
Mazhar, W.

#### **DERA ISMAIL KHAN**

Khan, J.

#### DOKRI, LARKANA, SINDH

Ali, M.

Khuhro, S.N. (Dr.) Rajput, S. (Dr.)

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# A Preliminary Study on Ichthyofaunal Diversity of Mir Kalam Dam, North Waziristan



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#### ABSTRACT

In the present study, Mir Kalam Dam in North Waziristan was explored for the first time to identify its ichthyodiversity. The study lasted for nine months, from February to October 2018 during which a total of 550 fish specimens were collected and identified as 11 species, belonging to three orders, three families, and nine genera. Among them Family Cyprinidae was the most speciose and the richest Family, represented by nine species i.e. Cyprinus carpio, Carassius auratus, Crossocheilus diplocheilus, Tor macrolepis, Puntius waageni, Aspidoparia morar, Barillius pakistanicus, Barillius vagra and Barillius bendelisis. The remaining two families were represented by only one species each. Family Siluridae was represented by Ompak pabda and Family Chandidae by Chanda nama. The most abundant fish in the Dam was Cyprinus carpio (26.18%) while the least abundant fish species were Tor macrolepis and Barillius bendelisis (0.36%) each. Results show that 27% species found are edible whereas the rest of the small fish species are important members of the food chains present in the Dam. Hence it can be concluded that the water of Mir Kalam Dam is productive and new species might be introduced in the Dam to provide economic support to the people of the area. Moreover, it is recommended that awareness projects should be conducted in North Waziristan for the ichthyo education in local people.

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**Authors' Contribution** 

FSW collected and identified the samples and made data arrangements. MFA prepared graphs, managed referencs and wrote the manuscript. ZH finalized identification, data analysis and discussion.

Key words
Fish diversity, Madi Khel Dam,
Cyprinidae, FATA, NWA

#### INTRODUCTION

The term Ichthyodiversity refers to the diversity of fish species based on different approaches (Haseeb et al., 2016). According to Bathe (2022), studies on the ichthyofaunal diversity and their protection in an aquatic ecosystem have long caught the attention of numerous fishery researchers. Fish are the most diverse and numerous vertebrate groups, occupying practically every hydrosphere niche, representing a wide variety of shapes, colors, and sizes. They make up more than half of all living vertebrate species and are classified into 515 families and 62 orders, totaling 27,977 species. In Pakistan, 193 freshwater fish species belong to the class Actinopterygii, subclass Teleostei, 3 cohorts, 6 super orders, 13 orders, 30 families, and 86 genera have been reported (Ali et al., 2020).

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Freshwater makes up around 3% of all water on Earth, whereas freshwater lakes and wetlands make up only 0.29 percent of the planet's freshwater. Nevertheless, they are home to approximately 10,500 fish species. Among them, 10,000 are exclusively freshwater fish (Mohapatra, 2022). Since freshwater fishes are the most diversified vertebrate group, they also have threatened species, and the decline is a global, regional, and local occurrence (Rehman et al., 2016). However, the alarming rate of species vanishing has been called the planet's sixth mass extinction (Mamilov et al., 2022). Therefore, efforts have been made to record the fish fauna of Mir Kalam Dam, North Waziristan. It is a part of Waziristan, a mountainous region in northwest Pakistan covering 4,473 sq. mi. Mir kalam dam, also known as Madi Khel dam, was executed by FATA Secretariat on a completion cost of 102.25 million Rupee. Geographically, the coordinates for the Dam are 33°00'05.2"N and 70°22'07.9"E. This Dam is a rain-filled type that utilizes the rainwater from Kharon Pal. It has a length of 695 ft. and a height of 52 ft. The dam command area is 375 Acres, and the catchment area is 22.70 Sq. Mi. The spillway capacity is 18,500 cusecs, and discharge is three cusecs.

#### MATERIALS AND METHODS

Study plan and fish sampling

The study plan was nine months, ranging from

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February to October 2018, to explore the freshwater fishes of district North Waziristan, Khyber Pakhtunkhwa, Pakistan. The samples were collected from the Madi Khel dam using several hooks and nets with variable mesh sizes. Fishes were preserved in 10 % formalin solution at the collection site, where live fish were immediately dropped into a large jar containing the solution. In contrast, large fish's belly and caudal peduncle region were injected with a 37 percent formaldehyde solution to protect it from internal viscera leakage and the outer body parts from infection caused by bacteria and fungi to prevent decomposition. Moreover, each jar was labeled with a serial number, collection date, and the collection site.

#### Identification of fish

The collected fish were transported to the Department of Zoology, University of Peshawar, Pakistan, where each specimen was characterized to the species level by considering different morphometric characters like dorsal, pectoral, pelvic, adipose, caudal, and anal fin rays. In addition, the lateral line scales, barbles, and teeth on the jaws and the proportion of the head to the body, the diameter of the eye, and its relationship to the head and total length of the fish was noted to employ various systemic and identification keys (Jayaram, 1999; Mirza and Sharif, 1982; Mirza and Sandhu, 2007; Talwar and Jhingran, 1991).

#### RESULTS AND DISCUSSION

During the nine months of 2018, a total of 550 fish specimens was collected from Madi Khel dam of District North Waziristan, Khyber Pakhtunkhwa, Pakistan, which were identified as 11 species (see Fig. 1) belonging to three orders, three families, and nine genera, as shown in (Table I). Among 550 samples, 445 (80.90%) samples belonged to the Family Cyprinidae, followed by 101 (18.36%) samples belonging to the Family Perciformes, while four (0.73%) samples belonged to the Family Siluridae, see Figure 2 and Table II. The present study reveals that the Family Cyprinidae is the most speciose and most prosperous Family, represented by nine species, i.e., Cyprinus carpio, Carassius auratus, Crossocheilus diplocheilus, Tor macrolepis, Puntius waageni, Aspidoparia morar, Barillius pakistanicus, Barillius vagra and Barillius bendelisis. In contrast, the remaining two families were represented by only one species, i.e., Family Siluridae was represented by Ompak pabda and Family Chandidae by Chanda nama. Moreover, results also demonstrate that the most abundant fish were Cyprinus carpio (26.18%), while the most diminutive abundant fish species were Tor macrolepis and Table I. Identified species with local names.

Barillius bendelisis (0.36%), (Fig. 3). The Madi Khel dam is home to both small and large fish, Barillius pakistanicus, Barillius vagra, Ompak pabda and Crossocheilus diplocheilus are the Dam's smaller fish. Given their small size, these smaller fish have no commercial value, but they serve a critical function in aquatic ecosystems because of their contribution to biodiversity. On the other hand, fish like Barilius pakistanicus are significant ornamental fish maintained in the aquariums to provide a fair commercial value (Ali et al., 2020). Furthermore, results show that 27% of species found are edible, like *Tor macrolepis*, Carrassius auratus, and Cyprinus carpio, as they are large fish with promising economic value to the local community. In the current study, Simpson diversity index 1-D was calculated as 0.82, representing good diversity. Similarly, the Shannon diversity index (H) was determined as 3.13, and with the help of that, we calculated the evenness for the data, which is 0.50. Additionally, the Z - A Index was determined as 0.25 as Hussain and Khan (2019) proposed to measure evenness.

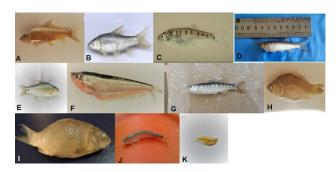


Fig. 1. Ichthyofaunal diversity in Madi Khel Dam, North Waziristan, KP, Pakistan.

A, Crossocheilus diplocheilus; B, Tor macrolepis; C, Barilius pakistanicus; D, Barilius vagra; E, Puntius waageni; F, Ompak pabda; G, Barilius bendelisis; H, Carassius auratus; I, Cyprinus carpio; J, Aspidoparia morar; K, Chanda nama.

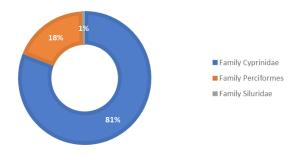


Fig. 2. Familywise ichthyofaunal diversity in Madi Khel Dam, North Waziristan.

S. No.	Order	Family	Genus	Species	Local Name
1	Cypriniformes	Cyprinidae	1. Crossocheilus	Crossocheilus diplocheilus	Butten
2			2. Tor	Tor macrolepis	Sunehri mahasher
3			3. Cyprinus	Cyprinus carpio	Gulfam
4			4. Carrassius	Carrassius auratus	Sunehri machli
5			5. Barillius	Barillius pakistanicus	Pakistani chilwa
6			6. Barillius	Barillius vagra	Lahori chilwa
7			7. Barillius	Barillius bendelisis	Patha chilwa
8			8. Puntius	Puntius waageni	Waageni popra
9			9. Aspidoparia	Aspidoparia morar	Gachal
10	Perciformes	Chandidae	10. Chanda	Chanda nama	Nama shisha
11	Siluriformes	Siluridae	11. Ompak	Ompak pabda	Mountain pafta

Table II. Nine-month collection and their total abundance.

S. No	Fish species	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Total	%
1	Crassocheilus diplocheilus	0	0	5	13	7	0	0	0	0	25	4.54%
2	Tor macrolepis	0	2	0	0	0	0	0	0	0	2	0.36%
3	Cyprinus carpio	17	9	5	3	11	33	42	13	11	144	26.18%
4	Carassius auratus	5	4	3	6	6	39	30	11	9	113	20.54%
5	Barillius pakistanicus	3	1	1	2	1	5	2	3	3	21	3.81%
6	Barillius vagra	1	1	0	1	0	0	1	0	0	4	0.72%
7	Barillius bendelisis	2	0	0	0	0	0	0	0	0	2	0.36%
8	Puntius waageni	1	0	3	0	0	7	3	28	7	49	8.90%
9	Aspidoparia morar	0	10	4	3	5	3	36	3	21	85	15.45%
10	Chanda nama	0	13	0	0	9	38	28	5	8	101	18.36%
11	Ompak pabda	0	0	0	0	0	0	1	3	0	4	0.72%
	Total	29	40	21	28	39	125	143	66	59	550	100

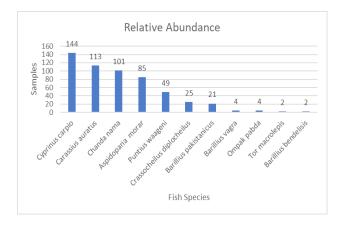


Fig. 3. Relative abundance of fish in Madi Khel Dam, North Waziristan.

Species identified in our study show little resemblance

with fish species reported from different sites of the same province. Like, Ali et al. (2020) has reported fourteen fish species from district Malakand, among them, Barilius pakistanicus, Barilius vagra, Carassius auratus, Ompak pabda, and Crossocheilus diplocheilus were the only fish species found similar to our catch. The absence of other species may be due to geographical differences and the water body nature, as in our case water was stagnant. Comparably, from river Panjkora, Dir upper, eleven fish species have been reported Among them, Carassius auratus and Crossocheilus diplocheilus were the only fish species similar to our findings (Liagat et al., 2014). Furthermore, Naveed et al. (2014) also reported ten fish species of the river Barandu, District Buner. Among them, Barilius pakistanicus was the only species found similar to our work. Again, differences in water bodies and terrain could explain this disparity.

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In contrast, the present study's icthyfaunal diversity bears some resemblance to the work of Ahmad (2018), who examined Murdar Algad in South Waziristan and discovered thirteen fish species. Five species were identified in our collection: Crossocheilus diplocheilus, Tor macrolepis, Barilius pakistanicus, Barilius bendelisis, and Barilius vagra. Other species, on the other hand, may be absent due to geographical differences and water bodies, as the water in our case was stagnant, whereas he worked in flowing water. Similarly, Rizwan (2018) reported twenty species from river Tank Zam, District Tank. Only seven species were identified in our study: Crossocheilus diplocheilus, Tor macrolepis, Barilius pakistanicus, Barilius bendelisis, Barilius vagra, Puntius waageni, and Ompak pabda. Once again, changes in water bodies and terrain may account for this discrepancy. Likewise, Haseeb et al. (2016) explored the ichthyofauna diversity of Tanda Dam, district Kohat and reported thirteen species among which Aspidoparia morar and Ompok pabda were identified in our study. Similarly, Zaryab et al. (2017) found seven species from Dandi Idhar Khel Lake, district Karak, and only Genus Aspidoparia and Genus Ompak were familiar from his work. Contrary, five fish species were reported from Dargai Pal Dam of South Waziristan among which Cyprinus carpio was related to our catch (Rehman et al., 2016). Likewise, the fish fauna of Dandy Dam, North Waziristan was also reported by Rehman et al. (2016) which includes five species of the Cyprinidae family. It demonstrates that the water of North Waziristan is favorable to Cyprinidae species, as in our case, Cyprinus carpio and Tor macrolepis were found comparative to his study.

#### CONCLUSIONS AND RECOMMENDATIONS

According to the findings of this study, Mir Kalam Dam North Waziristan has a large Cyprinidae family fauna, and the environmental circumstances in Mir Kalam Dam North Waziristan may be more beneficial for Cyprinidae family members. Other families, such as Perciformes and Siluriformes, are also supported. As a result, it is suggested that special augmentation programs should be implemented to begin utilizing fishery resources sustainably. Stocking natural water resources with a fish seed generated in hatcheries will be a frequent improvement. Additionally, the water bodies of North Waziristan are located in hilly locations, remote from populous areas, and are still free from significant pollution. If proper stocking and maintenance are taken, this water body can support a greater variety of fish species. During the rainy season, many fries, fingerlings, and adult fish are also washed away by overflowing water. Therefore, the

government should pay close attention to the reservoirs for fisheries growth. Furthermore, new fish species fingerlings should be introduced into the Dam to boost fish output and supply low-cost, high-quality protein to the locals.

Statement of conflict of interest

The authors have declared no conflict of interest.

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# Feasibility Assessment for the Salmo trutta fario (Brown Trout) and Oncorhynchus mykiss (Rainbow Trout) in Laspur and Lutkho Rivers in Chitral, Pakistan



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#### ABSTRACT

The current study was carried out to investigate the physicochemical parameters and assess the feasibility for the *Salmo trutta* fario (Brown trout) and *Oncorhynchus mykiss* (Rainbow trout) in Laspur and Lutkho rivers, District Chitral KP Pakistan. For this purpose, the physicochemical parameters such as pH, electric conductance, temperature, total dissolved solids (TDS), total hardness, alkalinity, and chlorine contents in the water samples collected from Laspur and Lutkho rivers, located in District Chitral using standard procedures were determined. The investigated values were compared with the standard values found earlier for optimum growth and productivity of *Salmo trutta* fario (Brown trout) and *Oncorhynchus mykiss* (Rainbow trout). In results, it was found that the average values of Laspur river were pH = 7.88, electric conductance = 0.447 mS cm<sup>-1</sup>, temperature = 11 °C, TDS = 63 mg L<sup>-1</sup>, hardness = 120 mg L<sup>-1</sup>, alkalinity = 74.3 mg L<sup>-1</sup> and Cl-1 = 11.16 mg L<sup>-1</sup> were recorded. Similarly, in case of river Lutkho, the average values regarding different physicochemical parameters were pH = 7.71, electric conductance = 0.448 mS cm<sup>-1</sup>, temperature = 8.5 °C, TDS = 61.5 mg L<sup>-1</sup>, hardness = 114 mg L<sup>-1</sup>, alkalinity = 104.3 mg L<sup>-1</sup> and Cl-1 = 9.83 mg L<sup>-1</sup>. In conclusion, the physicochemical parameters of both the rivers were slightly different from each other. Moreover, these parameters were found within the normal range for optimum growth and productivity of the studied trout species.

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#### Authors' Contribution

IA presented the concept. IA and AD curated data, planned methodology and administered the project. IA, AD, NUA, SB, RU and AJ did formal analysis. NUAsupervised the study. IA, NUA, SB, RU, AJ, IU, AD and MY validated the results. IA, IU, AD, and MY wrote the manuscript. IA, NUA, SB, RU, AJ, IU, AD and MY reviewed and edited the manuscript.

#### Key words

Physicochemical parameters, Rainbow trout, Brown trout, Water quality, Alkalinity

#### INTRODUCTION

Pakistan. Although, it contributes less to the GDP but earns foreign exchange through export. For the trout culture, a regular supply of clean quality water is required throughout the year. The higher production of trout fish can fulfill the requirements of white meat of the consumers and increase the economic level of the local people involved

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(Hassan *et al.*, 2007; Noroozrajabi *et al.*, 2013). Pakistan has many different climatic zones in a small area, which provide one of the best habitats for wild animals and fishes. Humans have a link from the very beginning with fishing which disturbed the quality and quantity of water (Altaf *et al.*, 2008).

Water quality is the analysis of physicochemical, biological, and microbiological parameters that affect the abiotic and biotic status of the ecosystem. The increase in the toxic materials disturb the water quality, not only affecting the health status of humans but also causing loss of oxygen, fish death, and aquatic biodiversity. For biodiversity and increased density, it is necessary to check regularly the water quality (Shedayi *et al.*, 2016; Ali *et al.*, 2019).

With the disturbance of pH and dissolved oxygen, the other parameters of water are affected directly or indirectly such as total dissolved solution, transparency, viscosity, and conductivity. The pH of water is affected 8 I. Ahmad et al.

by the concentration of dissolved ions salts and organic matter (Jabeen and Shedyi, 2011; Shedayi *et al.*, 2016).

Electric conductivity (EC) is to measure how much the solution passes electricity. It has a relation with salt contents as these are in ionic form in solution. Some of the salts are useful for fishes for osmoregulation in their body. Freshwater fishes are thriving at different EC levels. It may be used to give a rough estimate of the total dissolved solids (TDS) in water (Stone and Thomforde, 2004).

For fish raising, water quality and temperature should be optimum around the year. Temperature is one of the most important factors for aquatic life and the invertebrates are much sensitive to temperature. An adequate amount of clear water with a temperature range from 10-18 °C is required for the optimum growth and productivity of trout fish. Like the electric conductance, the TDS increase with an increase in temperature. It is also the reason that the values of TDS start to increase in the summer season (Khan et al., 2015; Shedayi et al., 2016; Hassan et al., 2007).

The high concentration of alkalinity in water is due to the presence of minerals salts such as carbonates and bicarbonates and such water is considered as hard. Alkalinity is high in summer due to the decomposition of organic matter. The concentration of chloride ions is different due to the presence of agriculture and domestic wastes. The presence of chlorides is due to the presence of the minerals, ions, and organic wastes (Khan *et al.*, 2015; Shedayi *et al.*, 2016).

Based on this discussion, it is necessary to maintain certain quality parameters at an optimum level for maximized production of fish. In addition, finding out the variation in natural prevailing conditions as under natural ecosystem, i.e. river is critical and helps to maintain those conditions under artificial ponds used for fish production. Based on these hypotheses, the present study was conducted to investigate the physicochemical parameters and assess the feasibility for the *Salmo trutta* fario (Brown trout) and *Oncorhynchus mykiss* (Rainbow trout) in Laspur and Lutkho rivers, District Chitral KP Pakistan

#### MATERIALS AND METHODS

Water sampling

Water samples were collected in clean plastic bottles; wash with the river water before collection of the water sample. Samples were taken from the running water of both rivers (Figs. 1 and 2) and were placed in a cooler having an ice bar to prevent the changes occurs during transportation to the Department of Zoology laboratory.

#### Physicochemical parameters

The physicochemical parameters of the natural

reservoir, Laspur, and Lutkho Rivers were recorded by using different methods and the available equipment in the laboratory. Some of the analyses were done on the spot such as temperature, pH, and electrical conductivity. The pH of the water was measured with the stick model Extech pH 110. The electric conductance was determined with a digital EC meter Taiwan made, I.D No. PLC/MBC (EnviR/009). The temperature of the water was measured with a digital centigrade thermometer at the sampling area by dipping in running water and waiting for five minutes. Total dissolved solids (TDS) of water samples were determined using a TDS meter (Digital instrument Taiwan made. I.D No. (Env. R/004).



Fig. 1. Laspur river.



Fig. 2. Lutkho river.

#### Total hardness

The titrimetric method was used to determine the total hardness of sample water. A conical flask was rinsed with sample water three times. 25 mL of sample water were taken and added 1 to 2 ml of buffer pH 10 (67.5g NH<sub>4</sub>Cl in 570ml conc. NH<sub>4</sub>OH), then 2-3 drops of Erichrome Black T indicator were added (0.5g sodium salt of 1-(1-hydroxy-2-naphthylazo)–5nitro-2naphthanol-4sulfonic acid dye in 100g Triethanolamine) and shake well. The solution was titrated with Ethylene diamine tetraacetic acid (EDTA) of 0.01 N until the color was changed to pink. The readings were noted directly and repeated three times.

$$Total\ hardness\ as\ {\rm CaCO_3}(mg/L)\ = \frac{V\times M\times 100\times 1000}{Sample\ volume}$$

Where V is the volume of EDTA used, M is the molarity of EDTA (0.01 M) and 100 is the molecular weight of  ${\rm CaCO_3}$ .

Table I. Physicochemical parameter of water samples from both the rivers.

Physicochemical parameters	Lutkho river	Laspur river
рН	7.80-7.95	7.50-7.85
Conductivity (mS cm <sup>-1</sup> )	0.432-0.461	0.434-0.462
Temperature (°C)	08-14	6-11
TDS (mg L <sup>-1</sup> )	288-295	286-296

Table II. Monthly variation in alkalinity.

Months	Alkalinity (mg L-1)			
	Lutkho river	Laspur river		
July	111.2	77		
August	97.2	63		
September	104.6	83		

Table III. Monthly variation in chlorides.

Months	Chloride (mg L <sup>-1</sup> )				
	Lutkho river	Laspur river			
July	10.9	11.4			
August	9.7	15			
September	8.9	07			

Table IV. River wise comparative values of calcium and magnesium hardness.

Physicochemical parameters	Lutkho river	Laspur river		
Calcium hardness	53 mg L <sup>-1</sup> CaCO <sub>3</sub>	54 mg L <sup>-1</sup> CaCO <sub>3</sub>		
Magnesium Hardness	61 mg L <sup>-1</sup> CaCO.	66 mg L <sup>-1</sup> CaCO.		

Alkalinity

The titrimetric method was used for the determination of Alkalinity. A water sample was taken in a conical flask, washed with distal water. Taken and washed with distal water. 25 ml of water sample was taken and three to four drops of methyl orange were added. It was titrated against 0.02 N H<sub>2</sub>SO<sub>4</sub> until the color of sample water was changed from orange to blue. The readings were noted directly from the burette and used in the following formula for total alkalinity calculations.

$$Total \ alkalinity \ as \ CaCO_3(mg/L) = \frac{N \times E \times V \times 1000}{Sample \ volume}$$

Where N is the normality of  $H_2SO_4$  (0.02 N), E is the equivalent weight of  $CaCO_3$  (50), and V is the volume of  $H_2SO_4$  (mL) used during titration.

Chlorides

The Argent metric method was used, in which the water sample was titrated against standard  $AgNO_3$  for the estimation of chloride. A total of 25 mL well-mixed water sample was diluted to 50 mL with distilled water.  $0.02~N~H_2SO_4$  were added to the same volume, as used for the total alkalinity followed by 2 to 4 drops of potassium chromate ( $K_2CrO_4$ ) indicator and titrated against  $0.0141~N~AgNO_3$  solution until the color changed to a pinkishyellow. The amount of chloride present was calculated from the amount of silver nitrate used as a titrant.

1 mL AgNO<sub>3</sub> = 0.45 mg chloride.  

$$Cl^{-}(mg/L) = \frac{A \times 1000}{Sample\ volume}$$

Where A is the volume of AgNO, used in titration

#### RESULTS AND DISCUSSION

Physicochemical parameter of water samples of both rivers

The physicochemical parameters are the most important external factors for the growth and development of fish. For an organism, environmental feasibility can be detected through physicochemical properties of water. The physicochemical parameters of both the rivers were within the normal range.

The physicochemical parameters of water were recorded in the course of study for both rivers (Table I). In case of Lutkho river, pH ranged from 7.80 to 7.95, electric conductance ranged from 0.432-0.461 mS cm<sup>-1</sup>, temperature ranged from 8-14 °C, TDS ranged from 288-295 mg L<sup>-1</sup>.

The physicochemical parameters of river Laspur were slightly different from that of river Lutkho (Table I). The pH for water samples were from 7.50 to 7.85, electrical conductance from 0.434 to 0.462 mS cm<sup>-1</sup>, temperature

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from 6 to 11C, and TDS from 286 to 296 mg L<sup>-1</sup>. Total hardness ranged from 53 to 61 mg L<sup>-1</sup> CaCo<sub>3</sub> of calcium and magnesium for river Lutkho and for river Laspur, it ranged from 54 to 66 mg L<sup>-1</sup> CaCo<sub>3</sub> of calcium and magnesium. Alkalinity and chloride levels were determined for three months. The alkalinity in July was 111.2 mg L<sup>-1</sup>, August 97.2 mg L<sup>-1</sup> and in September are 104.6 mg L<sup>-1</sup> for river Lutkho and for river Laspur in July was 77 mg L<sup>-1</sup>, August 63 mg L<sup>-1</sup>, and in September 83 mg L<sup>-1</sup>.

The chlorine values were also recorded for both rivers for three months. For river Lutkho in July 10.9 mg L<sup>-1</sup>, August 9.7 mg L<sup>-1</sup>, and in September 8.9 mg L<sup>-1</sup> were determined, and for river Laspur in July 11.4 mg L<sup>-1</sup>, in August 15 mg L<sup>-1</sup>, and on September 7 mg L<sup>-1</sup> are recorded.

Water quality plays an important role in fish production. If the physicochemical parameters of water quality are at the optimum level, the maximum production is obtained. The water quality and quantity affect biodiversity. The quality is influenced by climatic conditions (Ali *et al.*, 2019).

According to Petr (1999), Chitral and Kohistan are located in high mountain ranges with the temperate climatic region having clear cold water, which is suitable for the growth of trout fish.

The current physiochemical values of water such as pH, EC, Temp, TDS, alkalinity, and chlorine are almost according to Khan *et al.* (2015) with little differences. The differences and fluctuation in results may be due to environmental factors or by using different approaches.

#### **CONCLUSIONS**

The present study showed that physicochemical parameters of water from Lutkho and Laspur rivers of District Chitral have some ecological and climatic differences but the values were within the safe limits.

Statement of conflict of interest

The authors have declared no conflict of interest.

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# Parasitic Infestation in Fancy Birds: An Epidemiological Survey in Punjab

SIND BEX

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#### ABSTRACT

Fancy birds are reared as pet, sports, meat, recreational, teaching of taxidermy, biological and medical experimentations. Birds could be parasitized by several kinds of endo-parasites. Severe infections of these endo-parasites in birds significantly cause droopiness, loss of weight and diarrhea. Herein, parasites were identified through the feces by simple laboratory methods from selected districts of Punjab. Prevalence of the doves positive for the coccidiosis in Jhang was found (240/320) 75% and for Capillaria was (80/320) 25%. The incidence of the cockatiels birds positive for the coccidiosis in Layyah was (120/170) 70.5% and for Capillaria was (30/120) 17.6% while, mixed infection (Eimeria spp. + Capillaria spp.) prevalence was (20/120) 11.7%. Prevalence of the cockatiel and budgerigar identified for the parasite diagnosis in Jhang; both showed positive for the coccidian only and the prevalence of coccidiosis in cockatiels was (210/210) 100% and in the budgerigars was (120/120) 100%. The prevalence of the Pigeons bird positive for the coccidiosis in Muzaffargarh was found 70.7% and for Capillaria was 15.7%. No parasite was identified in (400) quail species from the Layyah district. This study represented that Coccidia and Capillaria were the most prevalent parasites found in the pigeons, doves and parrots species (cockatiels and budgerigars) in all the studied districts, but no parasite was found in the quails of Layyah district. This study recommends for the proper management and care of these birds' species in the studied districts to get risk of these parasitic infections to prevail these towards the other livestock populations and humans.

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MFQ supervised the entire work
and wrote manuscript. ARA and TH
Helped in planning of the experiment.
IL and ARA helped in the analysis and
refining the manuscript. IL helped in
statistical analysis of data. TH helped
in collection of data.

Key words
Fancy birds, Epidemiology, Parasitic infestation, Punjab

#### INTRODUCTION

Globally, the fancy birds play a crucial role in the societies and economy. These fancy birds may be peacock, pigeons, chicken, ducks, doves, lovebirds and parrots (Qamar et al., 2021). These are not only reared for gaming perseverance but also for fancy (ornamental) purpose (Akram et al., 2019). Several scientific reports have described that keeping animals as pet might bring positive effects on social as well as psychological wellbeing of humans. Moreover, for the treatment of major depressive disorder (MDD), social support is necessary that is not only provided by humans but also by different animals (Pereira and Fonte, 2018). Recently, the peoples

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prefer to keep fancy birds in captivity as pet instead of dogs and cats due to drastic intensification in human population and scarcity in accommodations (Akram *et al.*, 2019; Peng and Broom, 2021). These fancy birds are domesticated all over the world in the cities, villages and towns (Velden, 2019). However, these birds may be parasitized by several kinds of endo-parasites including cestodes, trematodes, acanthocephalans, nematodes, and protozoa (Waruiru *et al.*, 2018; El-Dakhly *et al.*, 2019). Therefore, proper management, nutrition and health care is essential for the prosperity of these bird species.

Nevertheless, these birds might act as the major source of infections closely associated with humans (Rahman *et al.*, 2020). Yet, these ornamental birds are usually ignored in terms of disease diagnosis and accompanying additional investigation (Qamar *et al.*, 2021). Acquaintance of heavy burden of parasites may lead to serious health problems or even may cause mortalities in exaggerated birds (Chacon *et al.*, 2021).

Therefore, the current study was performed to identify the fecal parasites of fancy birds in selected districts of Punjab through different laboratory methods. The testified prevalence of parasitic diseases would significantly help in the upgrading of potential control strategies. Knowledge about the analysis on the distributions of the diverse parasitic prevalence will support the clinicians to appropriately treat and provide awareness to the fanciers concerning appropriate control measures.

#### MATERIALS AND METHODS

Collection and examination of fecal samples

Fresh fecal samples different fancy birds were collected in the plastic bags from selected districts of Punjab. All the collected samples were analyzed through different parasitological techniques including direct smear inspection, sedimentation and flotation techniques.

For direct smear analysis, 0.5 g of fecal samples were mixed in 10 ml of normal saline. Following that, the debris was removed by filtering the suspension through a sieve. Then, a thin smear was made by the strained material (two drops) using glass slide and a coverslip. The slide was observed under the light microscope for the identification of oocysts (Deming *et al.*, 2008). The samples positive by direct smear investigation were further recognized through sedimentation and floatation methods.

Sedimentation method was done as proposed by (Latif *et al.*, 2016) in which 3 gs of fecal sample were mixed in distilled water and subsequently filtered. Thereafter, centrifugation of the filtrate was done for 2 min at 1500 rpm. Then, we discarded the supernatant and one drop of the sediments was placed on the clean glass slides to detect the parasitic oocysts under the light microscopy setup. The floatation method was carried out as proposed by (Faust, 1938). Counting of eggs per g (EPG) was done by means of McMaster method (Foreyt, 2013).

Statistical analysis

Statistical analysis was accomplished using Quantitative Parasitology tool (Reiczigel *et al.*, 2005). We calculated the prevalence values (percentage), mean abundance and mean intensity following previously employed statistical methods (Rózsa *et al.*, 2000).

#### RESULTS AND DISCUSSION

Prevalence of the doves positive for the *coccidiosis* in Jhang was found (240/320) 75% and for *Capillaria* was (80/320) 25%% in Jhang. No Mixed infection (*Eimeria* spp. + *Capillaria* spp.) prevalence was identified in doves. EPG counted through McMaster slide for the *Coccidia* was 700, while the EPG for *Capillaria* was 850 in Jhang of dove birds. The incidence of the cockatiels birds positive for the *coccidiosis* in Layyah found was (120/170) 70.5% and for *Capillaria* was (30/120) 17.6% while, mixed infection (*Eimeria* spp. + *Capillaria* spp.) prevalence was

(20/120) 11.7%. The incidence of budgerigars Layyah parrots observed was (80/90) 89% for coccidiosis and for Capillaria was (10/90) 11% and no mixed infection (Eimeria spp. + Capillaria spp.) was reported. Prevalence of the cockatiel and budgerigar identified for the parasite diagnosis in Jhang; both showed positive for the coccidian only and the prevalence of coccidiosis in cockatiels was (210/210) 100% and in the budgerigars was (120/120) 100%. EPG counted through McMaster slide for the Coccidia was 750, while the EPG for Capillaria was 700 in both cases of cockatiels and budgerigars of Layyah and EPG counted through McMaster slide for the Coccidia was 750 in both cases of cockatiels and budgerigars of Jhang. EPG count for the coccidian in both species of parrots was same in the both districts that showed very weak burden of the parasites. The prevalence of the Pigeons bird positive for the coccidiosis in Muzaffargarh was found 70.7% and for Capillaria was 15.7% in Muzaffargarh and the mixed infection prevalence was 13.4%. EPG counted for the Coccidia in Muzaffargarh was 750, while the EPG for Capillaria in Muzaffargarh was 700 in pigeons. No parasite was identified in (400) quail species from the Layyah district.

Different researchers have reported different prevalence rates of *coccidia* and *capillaria* spp. in fancy birds. Moreover, the prevalence and level of parasitic infection is influenced by the epidemiological factors such as sex, age, and breed (Djelmoudi *et al.*, 2014).

#### **CONCLUSION**

This study represented that *Coccidia* and *Capillaria* were the most prevalent parasites found in the pigeons, doves and parrots species (cockatiels and budgerigars) in all the studied districts, but no parasite was found in the quails of Layyah district. This study recommends for the proper management and care of these birds' species in the studied districts to get risk of these parasitic infections to prevail these towards the other livestock populations and humans.

Statement of conflict of interest

The authors have declared no conflict of interest.

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## Increasing the Digestibility of Corncob as Feed Ingredient by Urea Treatment for Grass Carp, *Ctenopharyngodon idella*



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#### ABSTRACT

The purpose of this study was to see how corncob treated with different doses of urea affected the growth of Ctenopharyngodon idella fingerlings. Treatment of corncob with graded levels of urea (0%, 2%, 4%, and 6 %, respectively) used in four experimental meals. Each of the four treatment groups (T0, T1, T2, and T3) included three replicates. The design of the study was completely randomized (CRD). Grass carp were fed with experimental feeds for 90 days at 3% body weight. Results revealed that final weight, weight gain, weight gain%, SGR, and FCR in T1 showed significant differences (P<0.05) from other treated groups. Feed intake was also significant (P<0.05) in T1. The survival rate of grass carp fingerlings was recorded the same in all the experimental groups. Significant (P<0.05) difference was observed in fish whole-body proximate as crude protein, dry matter and ash were high in T1. Crude fat contents were significantly high (P<0.05) in T0 while moisture content was found higher in T3 as compared to others. Hematological parameters showed significant (P<0.05) increased levels of white blood cells (WBC), hemoglobin (Hb), and hematocrit (HCT) in T1 and T2. ADC (dry matter) was significantly (P<0.05) high in T1 and T2. The crude fat digestibility was high in T3 but showed a non-significant (P>0.05) difference among other groups. In T1 crude protein, digestibility showed a significant (P<0.05) result. Amylase activity in T1 was significantly (P<0.05) high. Protease activity showed non-significant differences among treatments, while significantly (P<0.05) high lipase activity in T2. Water Physico-chemical parameters were found in optimal range throughout the experiment. It is concluded that corncob with urea treatment is an effective feed ingredient to improve growth, nutrient digestibility, activity of enzymes and hematological indices of grass carp.

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Authors' Contribution
SK, NK and MF planned the research design. SK, NK and KJI wrote the manuscript. SK, NK and MF studied growth, nutrient digestibility, proximate analysis, collected data, analyzed and interpret results. MNH helped in research work. MA, SN and SA assisted in statistical analysis and lab work.

Urea, Hematology, Digestive enzymes

Key words
Ctenopharyngodon idella, Corncob,

## INTRODUCTION

A quaculture dominates the production of aquatic animals and plants in Asia and worldwide (FAO, 2019). Asia is the major producer of aquatic food, produced about 102.9 million tonnes in 2017, making 91% of worldwide aquaculture production. Fish supplies more

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than 25% of animal protein to 1.25 billion people within 39 countries globally (FAO, 2018). Fish contains macronutrients including protein, omega-3 and omega-6 (PUFA), vitamins, carbohydrates as well as micronutrients like iron, copper, zinc, and selenium, presence of these nutrients make it balanced diet for human being (Mishra *et al.*, 2007; Tacon and Metian, 2013; Domingo, 2016).

Grass carp is herbivorous fish belonging to family Cyprinidae, inhabiting freshwater and culture all over the world (Tang *et al.*, 2013). It was first time introduced in Pakistan in1994 from China. It feeds on grasses as well as artificial diets that fulfill the requirements of this species (Hamza *et al.*, 2010). In intensive or semi-intensive culture Grass carp fed artificial feed, require protein content 300 to 400 g/kg and requirement of other energy sources such as dietary lipids 40g/kg and cannot utilized carbohydrates more than 400g/kg (Gao *et al.*, 2010). Like other countries

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in Pakistan carps culture at large scales (Aslam *et al.*, 2016). Over 5 million tonnes of grass carp is produced annually in the world (FAO, 2016), contributing mainly to freshwater aquaculture production about 5.1 million tonnes in 2013 (Pei *et al.*, 2015).

Artificial feed is the main component of aquaculture. The price of feed is directly related to protein based feed ingredients. The price of fish meal is increasing day by day, there is a need to use plant based protein sources in aqua feed (New and Wijkstrom, 2002). Carbohydrate containing feedstuff is cheap source of dietary energy (Krogdahl et al., 2005). Fish nutritionists and food producers pay more attention to culture fish species which utilize carbohydrates e.g., Labeo rohita and Ctenopharyngodon idella (Tian et al., 2010). As compared to other fish species Grass carp could digest and metabolize carbohydrates more efficiently (Gao et al., 2010).

Corn is individual from the oat grains, which produce dry, one-seeded products. Corncob is a waste produced from corn, estimated production of corn is 1.026 million tons globally (Chiellini et al., 2009). Corncob act as a potential feed ingredient in animal feed and act as feed substrate, which makes 27% to 30% of corn agrowaste (Kanengoni et al., 2015; Melekwe et al., 2016; Wachirapakorn et al., 2016). For the growth of various bacteria and fungi corncob is considered key substrate and production media for nutraceutical important enzymes and pharmaceuticals (Chapla et al., 2012). Much of the corncob is used for acetone-butanol (Kumar et al., 2014), carbon fuel cell (Jinshuai et al., 2014), and carbon adsorbent (Tsai et al., 2001). Utilization of corncob is less, due to high crude fiber content. Corncob contains nutrients like crude fiber 40.9%, non-nitrogen extract matter 52.5%, cellulose 12.05%, hemicellulose 28.06%, lignin 20.98%, crude protein 3.96%, water 7.08% and ash 1.1% (Tangenjaya and Wina, 2008). Fiber makes the digestion difficult along with it is beneficial to control the food in digestive tract that improves the digestion.

Alkali, dilute acid and organic solvents have been used in some pretreatments to degrade lignin and hemicellulose. Aqueous urea and urea in combination with other substances such as NaOH and choline chloride are used (Chen et al., 2015). Urea pretreatment has the potential to form reducing sugars from lignocellulosic material. Less developed countries are trying to improve animal feeding systems by urea supplementation. Urea supplementation is preferred as compared to other sources of ammonia because of being easily adaptable, cheap and less hazardous to use (Woyengo et al., 2004). This process improves the digestibility of the corn residues by reducing neutral detergent fiber and increasing the non-protein nitrogen content (Zanine et al., 2007). According to this

technique, urea dissolved in water and applied on the plant residues, ammonia gas will produce by this, which will break the bonds of lignin and hemicellulose and solubilize the hemicellulose (Rosa and Fadel, 2001).

Keeping in view the cheap and easily available carbohydrate (corncob) source, purpose of study was to determine the effect of urea treated corncob as feed ingredient on the growth, proximate composition, hematology, nutrient digestibility and enzyme activity of *C. idella*.

#### MATERIALS AND METHODS

The present trial was carried out in the fish seed rearing unit, Department of Fisheries and Aquaculture, University of Veterinary and Animal Sciences, Ravi Campus, Pattoki.

Experimental feed preparation

Corncob was collected from Pattoki, District Kasur, sun dried and then ground to a fine powder. Feed grade urea dissolved in water equivalent to 0%, 2%, 4% and 6% (w/v) was sprayed on the corncob and then mixed and stored at 24-26°C. After storage period, corncob plastic bags were exposed for two days for excess NH<sub>3</sub> to escape. Experimental feed formulation was given in Table I. All ingredients were fine ground, thoroughly mixed for 10-15 minutes with the addition of fish oil. 10-15% water was added to make dough for pellet formation. Laboratory pellet machine was used to prepare wet pellets. Air dried the pellets and stored in plastic bags. Experimental feed was designated as  $T_0$  (0%),  $T_1$  (2%),  $T_2$  (4%) and  $T_3$  (6%).

Table I. Formulation and ingredients composition of experimental feed.

<b>Feed ingredients</b>	T <sub>0</sub> (0%)	T <sub>1</sub> (2%)	T <sub>2</sub> (4%)	T <sub>3</sub> (6%)
Fish meal	25.0	25.0	25.0	25.0
Canola meal	15.0	15.0	15.0	15.0
Wheat flour	10.0	10.0	10.0	10.0
Urea treated corncob	40.0	40.0	40.0	40.0
Vitamin premix	2.0	2.0	2.0	2.0
Mineral mixture	2.0	2.0	2.0	2.0
Fish oil	5.5	5.5	5.5	5.5
Chromic oxide	0.5	0.5	0.5	0.5
Levels of urea	0.0	2.0	4.0	6.0

Experimental design and feeding management

Grass carp (*C. idella*) were used as experimental fish and acclimatized to indoor conditions for two weeks. Experimental design was completely randomized and each

treatment was triplicated. The trial was carried out in 12 glass aquariums having dimension (23x18x15 inches). Grass carp fingerlings having average body weight of 10-15g were randomly stocked in 12 aquaria with 15 fingerlings per aquarium. The fish were fed with experimental feed two times a day up to satiation (08:30am-05:30pm) for 90 days. Aerators were used to provide proper aeration. For determination of digestibility, fish were shifted into v-shaped digestibility tanks to collect the fecal matter for 15 days. During experiment water quality parameters like temperature (24.47 °C), pH (7.6-7.82), DO (6.56 ppm) and TDS (1444.9 ppm) were remained constant and monitored by using digital multimeters.

#### Calculations of fish growth parameters

At the time of initial stocking, fish wet body weight was measured individually. Growth parameters were calculated as follows:

Weight Gain (WG) = Final body weight (g) – Initial body weight (g)

FCR = Feed intake (g) / Wet weight gain (g)

SGR%/day= ln(w1)-In (w2)/number of days\*100

W1= initial wet body weight

W2= Final wet body weight

## Proximate analysis of fish, feed and feces

Four fish were taken from each replicate and were anaesthetized using (MS-222). Fish samples were dried in an oven at  $105\,^{\circ}\mathrm{C}$  for 14 h to determine the moisture content. Samples of fish, feed and feces in triplicates were dried, ground in pestle and mortar to make the fine powder. For further analysis, polythene bags were used to store powdered samples at room temperature. Proximate analysis was done by following the method of AOAC (2006). Crude protein (Nitrogen  $\times$  6.25) was calculated by the Kjeldahl apparatus. Crude ash of samples was assessed by using muffle furnace at 600  $^{\circ}\mathrm{C}$  for four to five h, while crude fat was measured using Soxhlet apparatus by ether extraction (Sr. No. 70861).

#### Digestibility

Feces of fish were collected from each digestibility tank after two h of feeding for 15 days. Sample of each group was placed into petri plates and dried in oven at 50°C and ground into powder. For apparent digestibility coefficients chromic oxide in feed and feces was measured by using spectrophotometer at 440 nm (Divakaran *et al.*, 2002).

## Digestive enzymes activity

After the completion of feeding trial, 4 to 5 fish from each aquarium were taken and anaesthetized with MS-222.

Intestine of 5 fish samples were collected and cold distilled water was used to wash. The intestine was homogenized and centrifuged for 10 min at 20,000 rpm. For assay of digestive enzymes supernatant was removed. Protease enzyme activity was assessed with Azocasein method (Garcia-Carreno, 1992). Lipase (Cherry and Crandell, 1932) and amylase activity was assessed by following the procedure of Bernfeld (1955).

#### Hematological analysis

For blood sampling five fish were randomly collected from each tank, a sterile syringe rinsed with EDTA, was used to take blood samples from non-anesthetized fish. Hematology parameters such as red blood cells, hematocrit, hemoglobin, white blood cells were determined by using automated hematology analyzer (MEK-655).

#### Statistical analysis

Data was subjected to SPSS (version 20) for one way ANOVA. Significant treatment means were compared through Duncan's multiple range test at 5% level of significance.

#### **RESULTS**

Growth

The effect of corncob treated with graded levels of urea on growth parameters is presented in Table II. Initial weight of *C. idella* fingerlings was kept nearly constant in all the experimental groups. The final weight, weight gain, weight gain%, SGR, FI and FE revealed a significant ( $P \le 0.05$ ) increase in T1 as compared to other treatments. T1 showed significantly ( $P \le 0.05$ ) low value of FCR. Survival rate did not show any change in all treated groups and non-significantly (P > 0.05) different.

## Fish whole body proximate composition

Proximate composition of *C. idella* fingerlings fed with experimental feed is given in Table III. Proximate composition of fish showed a significant ( $P \le 0.05$ ) difference. Dry matter and crude protein were found higher in T1 ( $19.43\pm0.20\%$ ) treated with 2% urea. Crude fat contents significantly higher ( $P \le 0.05$ ) in T0 decrease with the trend T3<T2<T1. Ash and moisture% were significantly ( $P \le 0.05$ ) high in T3 as compared to other treatments.

#### Nutrient digestibility

Nutrient digestibility showed significant ( $P \le 0.05$ ) results. Graded levels of urea in feed increased the absorption (digestibility) of dry matter significantly (P < 0.05) in T1, T2 as compared to T0 and T3. The crude

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fat digestibility % was found higher in T3, followed by T0, T2 and T1 but showed non-significant (P>0.05) difference among them. The crude protein digestibility % was significantly (P<0.05) improved in T1 (85.21±0.89) as compared to T2, T3 and T0 the lowest one (Table IV).

Hematological analysis of Ctenopharyngodon idella Among hematological parameters, the WBC, Hb and HCT showed significant (P<0.05) results whereas other indices were found non-significantly different after the addition of graded levels of urea (Table V). The graded levels of urea significantly (P<0.05) increase the number of WBC in T1 with the trend of T1>T3>T2 as compared to T0. T1 (8.67±0.05) showed significantly (P<0.05) higher Hb value fed 2% urea (T2). Higher HCT% was observed in T1 (35.46±0.01) as compared to other treatments. RBC showed non-significant (P>0.05) difference among T0, T2 and T3 except T1 (1.73±0.03).

Table II. Effect of urea treated corn cob on growth performance of *Ctenopharyngodon idella* fingerlings.

Parameters	T0	T <sub>1</sub>	T <sub>2</sub>	T <sub>3</sub>	p value
Initial weight (g)	$11.01 \pm 0.38^a$	$11.17 \pm 0.93^a$	$11.06\pm0.42^{a}$	10.90±1.21a	0.987
Final weight (g)	$35.51 \pm 0.49^{b}$	41.72±0.23°	33.22±0.41ª	$32.35 \pm 0.88^a$	0.000
Weight gain (g)	$24.50 \pm 0.25^{b}$	30.55±0.31°	22.16±0.13 <sup>a</sup>	$21.44 \pm 0.18^a$	0.000
Weight gain %	$223.06 \pm 8.03^a$	$274.90 \pm 16.08^{b}$	$200.87 \pm 8.36^a$	197.92±10.41a	0.004
SGR	$1.30\pm0.02^{a}$	$1.46 \pm 0.04^{b}$	$1.22\pm0.30^{a}$	$1.21\pm0.03^{a}$	0.004
FI	$40.54 \pm 0.35^a$	45.04±1.23°	$41.93 \pm 0.34^{b}$	$40.82 \pm 0.20^a$	0.000
FCR	$1.65\pm0.02^{b}$	$1.47\pm0.02^{a}$	$1.89\pm0.00^{c}$	$1.90\pm0.02^{c}$	0.000
SR %	98.40±0.60a	97.36±1.57a	97.10±0.36a	97.74±1.27 <sup>a</sup>	0.840
FE	60.44±1.05 <sup>b</sup>	67.83±1.02°	$52.85\pm0.10^{a}$	52.54±0.62a	0.000

Values (mean± SEM) in a row with unlike superscripts revealed significant (P≤0.05) difference. SGR, Specific growth rate; FI, Feed intake; FCR, Feed conversion ratio; SR, Survival rate; FE, Feed efficiency. For details of groups, see Table I.

Table III. Whole body proximate composition of Grass carp after 90 days feeding trial.

Parameters	$T_0$	T <sub>1</sub>	T <sub>2</sub>	T <sub>3</sub>	p Value
Moisture %	71.73±0.33 <sup>b</sup>	$70.26 \pm 0.39^a$	$72.35 \pm 0.02^{ab}$	73.03±0.40°	0.060
Dry matter%	$28.26 \pm 0.33^{b}$	$29.74 \pm 0.39^{c}$	$27.65{\pm}0.02^{ab}$	$26.96 \pm 0.40^a$	0.060
Ash%	$4.72\pm0.10^{b}$	$3.31 \pm 0.07^a$	$3.89 \pm 0.01^a$	$5.06 \pm 0.37^{b}$	0.001
Crude Protein%	$18.13 \pm 0.09^{b}$	$19.43 \pm 0.20^{\circ}$	$17.89 \pm 0.17^{b}$	$16.70 \pm 0.15^a$	0.000
Crude Fat%	$3.17 \pm 0.02^d$	$2.86\pm0.02^{\circ}$	2.73±0.01 <sup>b</sup>	2.65±0.01a	0.081

Values (mean± SEM) in a row with unlike superscripts revealed significant (P≤0.05) difference. For details of groups, see Table I.

Table IV. Effect of experimental feed on nutrient digestibility of grass carp.

Parameters	T0	T1	T2	Т3	p value
DM digestibility (%)	$73.34 \pm 0.54^a$	75.81±0.93°	$75.37 \pm 0.03b^{c}$	$73.60\pm0.41^{ab}$	0.038
CF digestibility (%)	$69.13 \pm 0.15^a$	$66.60\pm2.86^a$	$67.90 \pm 1.67^a$	$72.56\pm5.10^{a}$	0.574
CP digestibility (%)	$82.28 \pm 0.06^a$	$85.21 \pm 0.89^{b}$	$83.23 \pm 0.54^a$	$82.81 \pm 0.32^a$	0.026

Values (mean± SEM) in a row with unlike superscripts revealed significant (P≤0.05) difference. DM, Dry matter; CR, Crude fat; CP, Crude protein. For details of groups, see Table I.

Table V. Effect of urea treated corn cob on hematological parameters of grass carp.

Parameters	Т0	T <sub>1</sub>	T <sub>2</sub>	T <sub>3</sub>	p Value
RBC (10 <sup>6</sup> /μL)	$2.27 \pm 0.26^{b}$	$1.73\pm0.03^{a}$	$2.15 \pm 0.02^{ab}$	$2.18\pm0.02^{b}$	0.079
WBC $(10^3/\mu L)$	16.37±0.01a	$31.43 \pm 0.65^d$	$25.23 \pm 0.06^{b}$	$27.73\pm0.49^{c}$	0.000
Hb (g/dL)	$7.14 \pm 0.15^a$	$8.67 \pm 0.05^{b}$	$7.21 \pm 0.24^{a}$	$7.02\pm0.19^{a}$	0.001
HCT (%)	28.24±0.02a	35.46±0.01°	28.33±0.01a	$34.48 \pm 0.31^{b}$	0.000

Values (mean $\pm$  SEM) in a row with unlike superscripts revealed significant (P $\le$ 0.05) difference. RBC, Red blood cells; WBC, White blood cells; Hb, Hemoglobin; HCT, Hematocrit. For details of groups, see Table I.

Table VI. Effect of urea treated corn cob on digestive enzymes activity of grass carp.

Parameters	T0	T <sub>1</sub>	T <sub>2</sub>	$T_3$	p value
Protease (U mg-1)	$5.27 \pm 0.26^a$	$5.96\pm0.16^{a}$	$6.38\pm0.41^{a}$	$5.57 \pm 0.76^a$	0.407
Lipase (U mg-1)	$1.82 \pm 0.27^{b}$	$2.05{\pm}0.35^{ab}$	$2.28 \pm 0.26^{b}$	$1.92\pm0.23^{a}$	0.668
Amylase (U mg-1)	$14.42 \pm 0.15^a$	$16.61\pm0.67^{b}$	$13.40\pm0.51^a$	$13.66 \pm 0.56^{a}$	0.009

Values (mean± SEM) in a row with unlike superscripts revealed significant (P≤0.05) difference. For details of groups, see Table I.

Digestive enzymes activity

Fish feeding with 2% level of urea treated corncob based feed (T1) indicated increase in amylase activity (16.61±0.67) significantly (P<0.05) in Grass carp. The protease enzymes showed non-significant differences among treatments while lipase activity in T2 (2.28±0.26) found significantly higher followed by T1 (2.05±0.35) compared to T0 (1.82±0.27) (Table VI).

#### **DISCUSSION**

In present study, corncob treated with different levels of urea found effective in the growth performance of C. idella fingerlings. The weight gain was high in T1 (30.55±0.31g), which was fed with the diet supplemented at 2 % urea and is similar with the outcomes of Klahan et al. (2016) who fed nile tilapia with different levels of corncob for 120 days. Jawdhari et al. (2020) reported that common carp showed increase in weight gain when it was fed a diet which was replaced by corn residues. On the contrary, high supplementation of urea in the diet decreases the weight gain (Olson et al., 1999). Hammed et al. (2003) reported higher SGR in Catla catla and C. idella when fed with diet based on plant residues. FCR of our study showed better results in T2 (1.89) and T3 (1.90) which was in accordance with Klahan et al. (2016). The feed intake in current study revealed that fish showed improved feed intake (45.04) fed at 2% urea which was in line with Aruwayo (2018) who reported that increase in urea level above 2% reduced feed intake. Survival rate remained same in all the treatment groups about (97%) inline with the results of Ren et al. (2015) who found that survival rate (96%) of snout bream fed with carbohydrate base sources of feed.

The inclusion of 2% level of urea in feed revealed better results for proximate composition of fish during current study like crude protein showed significant differences. Higher % of ash was observed in T1 showed significant (P>0.05) difference among treated groups and are in line with Metwally and Gellal (2009) who observed that ash and dry matter of fish showed significant differences when fed on remaining plant materials.

The health of fish is assessed via hematological analysis. Results of RBC and Hb of T0 without urea

treatment and T1 treated with 2% urea were same and in line with (Klahan et al., 2016) who stated RBC and Hb were non-significantly (P>0.05) different when tilapia fed with graded levels of corncob and the hematocrit was not according to the results previously reported. The present study agreed with the results of Daramola et al. (2005) who observed hemoglobin in range 5.98-9.22 g/dl respectively as compared to our results where T1 had the highest value of hemoglobin (8.65 g/dL) which is important indicator of increased oxygen carrying capacity in fish.

In the present study, urea treated corncob affects the digestive enzymes activity. Amylase enzyme showed significantly high activity (16.61 mg/min/ml) in T1, similar with the results of Aslam et al. (2018) who stated that amylase concentration was high in C. idella fed feed with 21% duckweed when cultured the fish in monoculture and polyculture system. This improvement may be related with the enzymatic digestion of feed which in turn improved the feed digestion. The digestive processes of fish are not as well understood as those of other vertebrates, although digestive enzyme investigations in most vertebrates demonstrated a similar tendency. The majority of research on digestive, proteolytic, and amylase enzyme activity revealed that various species can utilize carbohydrates and proteins in different ways (Hidalgo et al., 1999). Studies on herbivorous and omnivorous fishes showed they utilize carbohydrate efficiently than carnivorous fishes (Feranadez et al., 2001; Drewe et al., 2004; Horn et al., 2006).

Chromic oxide is most widely used marker for nutrient digestibility in fish i.e., tilapia, rainbow trout, rohu and grass carp. Dry matter digestibility increased when urea added in the diet due to the loose lignin-cellulose bound as a result of the ammoniation and fermentation process that predigest the complex compounds. Prastyawan *et al.* (2012) stated that the dry matter digestibility of corncobs treated with ammonia fermentation would increase the level of starter. In our study improved dry matter digestibility and protein digestibility was observed in T1 (75.81%) treated with 2% urea that was in line with the findings of Yulistiani *et al.* (2011) who stated that incorporating 2% urea into the corncobs could increase dry matter digestibility by 31%. Our results are different with Fitria *et al.* (2020) who stated that dry matter digestibility increased in animals at

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5% level of urea. In this study crude protein digestibility of grass carp showed significant results as T1 (85.21%) was significantly different from the other groups and were similar to the value reported by Hamad *et al.* (2011) in the study of *C. catla* and *C. idella* fed with different levels of urea. This improvement in nutrient digestibility would ultimately increase growth rate of fish.

The Physico-chemical parameters of water were in optimal range. Our results were in line with Hammed *et al.* (2003) who reported there was no change in water quality parameters when treated with urea for rearing of catfish but DO was not in accordance with Aniyikaiye *et al.* (2019) who stated that oxygen concentration in water is not constant.

#### **CONCLUSION**

The present study revealed that corncob treated with 2% of urea significantly enhanced the growth performance, whole body composition, digestive enzymes activity, nutrient digestibility and showed no negative effect on hematological indices. Urea level above 2% did not give better results that might be its pungent smell to affect feed intake.

Statement of conflict of interest

The authors have declared no conflict of interest.

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## **CITATIONS**

## Prof. Dr. Muzaffer Ahmad Lifetime Achievement Award 2021

Prof. Dr. Mohammad Perwaiz Iqbal, Sitara-i-Imtiaz Life Sciences Department, University of Management and Technology, Lahore



Professor Iqbal obtained his M.Sc. Chemistry from the Government College, Lahore in 1970 with distinction. He then joined the University of Maryland's International Center for Medical Research and Training, Lahore, where he worked on genetics of various mosquito species in Pakistan. In 1974, he went to USA and joined the New York Medical College where he started his pioneering work on folate metabolism under the mentorship of Professor Sheldon Rothenberg. This novel research work led to the development of first radioimmunoassay for dihydrofolate reductase (DHFR), a key enzyme in cell proliferation. In 1976 he joined the New York University for his Masters and PhD degrees in Biological Sciences in 1981. He was appointed as faculty in the Downstate Medical Center, State University of New York, Brooklyn and was soon promoted to the rank of Assistant Professor. During this period, he developed novel radioassays for dihydrofolic acid and the anti-cancer drug, Methotrexate which had immense application in cancer chemotherapy. In 1983, he joined the Aga Khan University as founding faculty in Biochemistry and became Professor in 1992. He served the University for 36 years with distinction. He has received three Outstanding Teacher Awards (1995, 2006, and 2010) and an Award of Excellence in Education in 2006. He retired in 2019 and was bestowed upon the title of Professor Emeritus in 2019. He has published 170 research papers, received numerous research grants worth millions of rupees and provided guidance to numerous PhD students. He has been a recipient of several other awards including INFAO Foundation Gold Medal in Biological Sciences (1992), PAS/ INFAQ Gold Medal in Medical Sciences (2001). He is also Fellow of Pakistan Academy of Medical Sciences, Pakistan Academy of Sciences, Pakistan Society for Biochemistry and Molecular Biology and Zoological Society of Pakistan. He was decorated with the Civil Award, Sitara-i-Imtiaz in 2002 by the President of Pakistan for his outstanding contribution to teaching and research in Pakistan.

Prof. Dr. Shahid Mahmood Baig, Sitara-i-Imtiaz Chairman, Pakistan Science Foundation, Islamabad



Dr. Shahid Mahmood Baig completed his MSc and MPhil in Biology in 1984 and 1986, from Quaid-i-Azam University, Islamabad. He joined NORI -PAEC as a Research Scholar in 1986 and as a Scientific Officer in 1987. He was transferred to NIBGE in 1995 and completed his PhD from Bosphorus University Turkey and Quaid-i-Azam University Islamabad in 1996. Dr. Baig completed his postdoctoral fellowship from Duke University USA in 2012. He served as Deputy Chief Scientist, Group Leader and Head of Health Biotechnology Division, NIBGE -PAEC and Professor at Aga Khan University Karachi. Currently he is working as Chairman Pakistan Science Foundation. Dr. Baig established world class research facilities for Human Disease Gene Identification, prenatal diagnosis and prevention of inherited diseases in Pakistan. The team led by him has identified 6 novel genes out of 25 known so for causing primary microcephaly and involved in human brain development published in Nature Genetics, Nature Communication, Journal of Clinical Investigation, Annals of Neurology, American Journal of Human Genetics and Human Molecular Genetics. He has characterized molecular genetic defects in >3,000 large consanguineous Pakistani families with various inherited diseases using state of the art next generation sequencing (NGS) for disease prevention and precision medicine. He as also provided mutation screening, cascade testing and genetic counseling to >2000 families having at least one thalassemia major child, and provided retrospective first trimester prenatal diagnoses in 1500 pregnancies at risk of thalassemia major. He has established very strong research and academic linkages with Universities and research institutions in Sweden, Denmark, Germany, Switzerland, France, Norway, UK, USA, and China. Dr. Baig has authored 131 peer reviewed publications and produced 31 PhD scholars.

Prof. Dr. Syed Shamim Ahmed, CEO, Biomedicine & Biosciences Consultancy, Karachi.



Prof. Dr. Sved Shamim Ahmad is Visiting Research Professor in Greenwich University, Karachi and Consultant Editor of Greenwich Research Journals on Humanities and Business Studies. He has previously held positions of Research Professor in Karakorum International University (KIU), Gilgit, HEC Eminent Professor in University of Karachi, Member Sciences on Syndicates & Senates of Shah Abdul Latif University & University of Sindh, World Health Organization (WHO) International Long -Term Consultant in South & SE-Asia, Research Professor in McGill University, Montreal, Canada, Senior Fulbright Research Professor at University of California, Davis, USA. Research Professor, Toyama Medical & Pharmaceutical University, Japan, Senior Research Fellow at American University of Beirut (AUB) and Christies Hospital & Holt Radium I nstitute, Manchester, UK, Senior Experimental Research Officer in University of Salford, UK, Commonwealth Research Fellow in Liverpool School of Tropical Medicine, UK and Professor & Chairman, Department of Zoology, Sindh University, Jamshoro. He has vast experience of human parasitic tropical diseases, animals studies on Biology, Ecology & Biological Control of vectors of public health importance and endemic bio-medical research issues of Pakistan. Besides that Prof. Ahmed has developed several on-Line courses for medical professionals.

Prof. Dr. Juma Khan Kakar Department of Zoology, University of Balochistan, Quetta.



Dr. Juma Khan Kakar obtained his BSc from Aligarh Muslim University in 1968, M.Sc Zoology in 1985, M. Phil in 1992, and Ph.D. in the field of Parasitology in 2002 from University of Balochistan, Quetta. He worked on Phlebotomine sand flies and cutaneous leishmaniasis in Balochistan for his doctoral degree, He spent time on the subject of his interest in the laboratories of Prof. Dr. Yoshihisa Hashiguchi of Kochi University, Japan, Prof. Dr. Robert Killick - Kendrick in Imperial College, Ascot, England, Prof. David Evans in London School of Hygiene & Tropical Medicine, and Dr. Richard Lane in British Museum Natural History London, and made significant contributions in the field of sand flies and leishmaniasis. He has published about 107 research papers on sand flies and leishmaniasis, and supervised three Ph.Ds.

Prof. Dr. Muhammad Akhtar, Ex-Chairman, Department of Zoology, University of the Punjab, Lahore



Prof. Dr. Muhammad Akhtar obtained his M.Sc. and Ph.D. degree from University of the Punjab, Lahore. He joined Department of Zoology, University of the Punjab as Research Officer 1982 and retired as Professor of Zoology in 2015. He is currently working as Professor of Zoology in University of Central Punjab, Lahore. Prof. Akhtar headed Department of Zoology, University of the Puniab from 2004-2015 and also worked as Registrar University of the Punjab from 2010 to 2012. Prof. Dr. Akhtar spent his entire academic career in promoting the subject of Palaeontology. He established Dr. Abu Bakr Fossil Display and Research Centre in March 2015 to pay tribute to Dr. Abu Bakr, the pioneer Vertebrate Palaeontologist of Pakistan. Prof. Dr. Muzaffar Ahmed had strong desire in his life for the construction of new building of Zoological Museum in the Department of Zoology, he succeeded to establish Zoological Museum in the tenure of Prof. Akhtar in 2015. Prof. Akhtar reported 15 mammalian new extinct species from Siwalik Hills of Pakistan. His 207 research publications in national and international impact factor journals and 21 Ph.Ds. in Mammalian Palaeontology speak for his lifelong commitment to the subject.

Prof. Dr. Nusrat Jahan, Ex-Chairperson, Department of Zoology, Government College University, Lahore.



Prof Dr. Nusrat Jahan obtained her Ph.D from School of Life Sciences. University of Keele, Staffordshire, UK in 1999. After her stay in University of Keele for postdoctoral work she proceeded to University of Florida, Gainesville (2001, 2002) and University of Georgia, Athens, USA in 2002 to gain further experience in the field of Medical and Molecular Entomology. She has 40 years of research and teaching experience. Her major contributions have been in biological and integrated management of vectors involved in malaria and dengue and genetic basis of public health diseases. Dr. Jahan has also contributed to studies on genetic association and environmental risk factors regarding Diabetes type 2, Myocardial Infarction, Asthma and female reproductive dysfunction. These findings will help to develop better diagnosis and treatment of their diseases in Pakistan. Dr. Jahan established an Animal Tissue Culture Laboratory for Wolbachia-based dengue control strategy in Pakistan. Dr. Jahan was awarded Best University Teacher Award 2012-2013 by HEC Islamabad, two Research Productivity Awards in 2015 and 2016 by Pakistan Council for Science and Technology, Islamabad. Dr. Jahan has 67 research publications and supervised 11 PhD scholars.

## **Zoologist of the Year Award 2020**

Prof. Dr. Farah Rauf Shakoori, Department of Zoology, University of the Punjab, Lahore.



Prof. Dr. Farah Rauf Shakoori obtained her Master's degree in Zoology from University of the Punjab in 1989, and later proceeded to University of Massachusetts Medical Center and Comprehensive Cancer Center, Worcester, USA to undertake her Ph.D. program in collaboration with the University of the Punjab. During her stay in America, she involved herself in understanding molecular mechanism of cell cycle control and abnormal cell growth control which is observed in cancer related diseases. She has published 92 original articles in elite scientific journals including Nature, PNAS USA, JCB, Journal of Cellular Physiology, USA, Molecular Biology Reports, Biochemistry, Journal of Cellular Biochemistry, she has accumulated an impact factor of 117.01, H-index 22, and i10-index 38, and total citations is 1312 according to Google scholar. She has contributed a chapter "Numerical changes in chromosomes" in a book "Chromosome structure and Aberrations" published by Springer Verlag. Dr. Farah Shakoori is engaged in research project in the areas of Environmental Biotechnology and Molecular Biology. On the basis of scientific achievements, Dr. Shakoori was awarded Research Productivity Allowance by PCST for the Years 2002, 2011 and 2012. She has been listed in Productive Scientists of Pakistan by PCST since 2007. She was awarded Prof. Dr. Mirza Azhar Beg Gold Medal in 2005 and Prof A. R. Shakoori Gold Medal in 2008 by the Zoological Society of Pakistan. She has also won Open Gold Medal in Zoology of Pakistan Academy of Sciences in 2014 on the basis of outstanding research contribution in the field of Molecular Biology and Environmental Biotechnology. She is a regular member of international Organization for women in science for the developing world (OWSDW) under the umbrella of TWAS.

<sup>\*</sup>Other applicants of this award were. Prof. Dr. Mukhtiar A. Mahar, Prof. Dr. Abdul Rehman, Dr. Muhammad Sarwar, Dr. Noor-un-Nisa, Dr. Muhammad Ather Rafi, Dr. Javed Ahmed Ujan and Dr. Zulfiqar Ali

## Prof. Dr. A.R. Shakoori Gold Medal 2020

Dr. Muhammad Sajid Hamid Akash, Chairman, Department of Pharmaceutical Chemistry, Government College University, Faisalabad.



Dr. Muhammad Sajid Hamid Akash is currently working as an Associate Professor and Chairman Department of Pharmaceutical Chemistry, Government College University Faisalabad (GCUF), Pakistan. He did his PhD in 2015 from Zhe jiang University, China. During his PhD, he worked on the role of inflammatory mechanisms in pathogenesis and development of diabetes mellitus and its treatment strategies. He has won 4 research grants from Higher Education Commission (HEC) of Pakistan and one research from GCUF. Till now, he has published 146 articles with cumulative Impact Factor 450 and 4700 citations with h-index 35. He has published 4 books in Springer Nature and 1 book in Elsevier. Moreover, he has also published 50 book chapters with international publishers like Springer Nature, Elsevier, Material Research Forum LLC, and CRC Press. Currently, he is also working as "Associate editor" in "BMC Endocrine Disorders", "PLOS ONE", "Biochemical Genetics", "Artificial Cells, Nanomedicine and Biotechnology" and "Endocrine, Metabolic & Immune Disorders - Drug Targets". Based on his significant scientific contributions in the field of Pharmaceutical Sciences, he was awarded "Research Productivity Award" by Pakistan Council for Science and Technology (PCST) in 2014 (Category C), 2015 (Category B), 2016 (Category B) and 2017 (Category C). Based on his outstanding scientific contributions, Pakistan Academy of Sciences awarded him the most prestigious award "PAS Gold Medal" in the field of Health Sciences in 2019.

<sup>\*</sup>Other applicants of this award were. Dr. Muhammad Zafar Hashmi, Dr. Kanwal Rehman, Dr. Saadullah Khan, Dr. Muhammad Khan, Dr. Asif Nadeem, Dr. Wali Khan and Dr. Muzafar Shah

## Prof. Imtiaz Ahmad Gold Medal 2020

Dr. Muhammad Irfan Ullah Department of Entomology, College of Agriculture, University of Sargodha, Sargodha



Dr. Muhammad Irfan Ullah obtained M.Sc. (Hons.) in Entomology from University of Agriculture, Faisalabad, Pakistan in 2003 and earned his Ph.D. from University of Nebraska -Lincoln, USA in 2012. He presently is working as an Assistant Professor at Department of Entomology, University of Sargodha. For his Ph.D. research, he worked on the aspects of insect plant interaction, insect stress biology and insect genetics. He has established a laboratory of biological control in collaboration with China where he is working on management of citrus insect pests. Dr. Irfan has published more than 70 research papers in the journals of international repute published 01 Book on Insect Behavior and several book chapters. He has supervised 03 PhD theses.

<sup>\*</sup>Other applicant of this award was Prof. Dr. Hafiz Muhammad Tahir

## Prof. Dr. Nasima M. Tirmizi Gold Medal 2020

Dr. Sher Khan Panhwar
Centre of Excellence in Marine Biology,
College of Agriculture,
University of Sargodha,
University of Karachi,
Karachi.



Dr. Sher Khan Panhwar obtained MSc from University of Sindh, Jamshoro in 2001, M.Phil f rom CEMB, University of Karachi in 2007, Ph.D. from Ocean University of China in 2012, postdoctoral research from Zhejiang Ocean University in Fisheries Management, besides several trainings in China and Pakistan. During PhD research, he worked on fish population dynamics and fisheries resources of Pakistan using advanced fisheries models and approaches during postdoctoral research work at Zhejiang Ocean University worked on conservation and management of fish stocks of the East China Sea (Zhejiang). Dr. Panhwar was appointed as Lecturer in 2010 and upgraded as Assistant Professor in 2012 at the Centre of Excellence in Marine Biology, University of Karachi. Dr. Panhwar has published 40 research articles in internationally reputed journals, supervised 2 Ph.D. theses and completed five national and international research projects.

<sup>\*</sup>Other applicant of this award was Dr. Noor Us Saher

# RECIPIENTS OF GOLD MEDALS AWARDED BY THE ZOOLOGICAL SOCIETY OF PAKISTAN

## 1. Muzaffer Ahmad Gold Medal 2020

The 27<sup>th</sup> Muzaffer Ahmad Gold Medal 2020 was received by Ms. Maham Arif a student of University of the Punjab, Lahore for standing first in the recent M.Sc. Zoology examination.



Ms. Maham Arif

## 2. Afsar Mian Gold Medal 2020

The 12<sup>th</sup> Afsar Mian Gold Medal 2020 was given to Ms. Sameen Fatima a student of the Arid Agriculture University, Rawalpindi for standing first in the recent M.Sc. Biology/Zoology examination.



Ms. Sameen Fatima



## 3. Muhammad Afzal Hussain Qadri Memorial Gold Medal 2020

The 23<sup>rd</sup> Muhammad Afzal Hussain Qadri Memorial Gold Medal 2020 was awarded to Mariya Aziz student of Karachi University for standing first in the recent M.Sc. Zoology examination.

## 4. Prof. Dr. S.N.H. Naqvi Gold Medal 2020

The 16<sup>th</sup> Prof. Dr. S.N.H. Naqvi Gold Medal 2020 was given to Dr. Hina Manzoor for obtaining Ph.D. degree in Zoology with specialization in the field of Toxicology from University of Karachi.



Dr. Hina Manzoor

## 5. M.A.H. Qadri Memorial Gold Medal 2019

The 20<sup>th</sup> M.A.H. Qadri Memorial Gold Medal 2020 was given to Dr. Bushra Siyal for obtaining Ph.D. degree in Zoology with specialization in the field of Parasitology from University of Karachi.



Dr. Bushra Siyal

## 6. Mujib Memorial Gold Medal 2020

The 27<sup>th</sup> Mujib Memorial Gold Medal 2020 was given to Sana Riafat a student of Karachi University for standing first in the recent M.Sc. Zoology examination with specialization in Parasitology.



Ms. Sana Riafat

## 7. Prof. Dr. Muhammad Ali Gold Medal 2020

The 4<sup>th</sup> Prof. Dr. Muhammad Ali Gold Medal 2020 was awarded to Tayba Ameen a student of Government College University, Faisalabad for standing first in the recent M.Sc. Zoology examination.



Ms. Tayba Ameen



## 8. Prof. Dr. Syed Iftikhar Hussain Jafri Gold Medal 2020

The 4<sup>th</sup> Prof. Dr. Syed Iftikhar Hussain Jafri Gold Medal 2020 was awarded to Nazia Nawab a student of University of Sindh, Jamshoro for standing first in the recent final B.S. Examination of Freshwater Biology & Fisheries.



Ms. Nazia Nawab

## 9. Ahmed Mohiuddin Memorial Gold Medal 2020

The 15<sup>th</sup> Ahmed Mohiuddin Memorial Gold Medal 2020 was awarded to Ms. Khushboo a student of University of Sindh, Jamshoro for standing first in the recent M.Sc. Zoology examination.



Ms. Khushboo



The 7<sup>th</sup> Prof. Dr. S.S. Akbar Memorial Gold Medal 2020 was awarded to Mr. Jaffer Hussain a student of University of Sindh, Jamshoro for standing first in the recent M.Sc. Zoology examination with specialization in Entomology.



Mr. Jaffer Hussain

## 11. Prof. Dr. Muhammad Saeed Wagon Gold Medal 2020

The 2<sup>nd</sup> Prof. Dr. Muhammad Saeed Wagon Gold Medal 2020 was awarded to Ms. Maryam a student of University of Sindh, Jamshoro for standing first in the recent BS-IV Zoology examination.



Ms. Maryam



## 12. Prof. Dr. Naeem Tariq Narejo Gold Medal 2020

Prof. Dr. Naeem Tariq Narejo Gold Medal 2020 was awarded to Mr. Kamran Ali Abro a student of University of Sindh, Jamshoro for standing first in the recent M.Sc. Fresh Water Biology & Fisheries examination.



Mr. Kamran Ali Abro

## SOME GLIMPSES OF ACADEMIC SESSIONS AND THE CONGRESS PARTICIPANTS



The Vice Chancellor receiving Prof. Dr. Muhammad Iqbal Chaudhary, the Chief Guest



Inaugural session: Left to right, Dr. Abdullah G. Arijo, Dr. Farooq Hassan, Dr. M. Iqbal Chaudhary, Dr. Fateh Muhammad Marri, Dr. Abdul Rauf Shakoori, Mr. Abdul Aziz Khan



Left to right: Dr. Abdullah G. Arijo, Dr. Farooq Hassan Dr. M. Iqbal Chaudhary



Left to right: Dr. M. Iqbal Chaudhary, Dr. Fateh Muhammad Marri, Dr. Abdul Rauf Shakoori



Left to right: Dr. Abdullah G. Arijo, Dr. Farooq Hassan, Dr. M. Iqbal Chaudhary, Dr. Fateh Muhammad Marri, Dr. Abdul Rauf Shakoori, Mr. Abdul Aziz Khan



Participants standing in honour of National Anthem



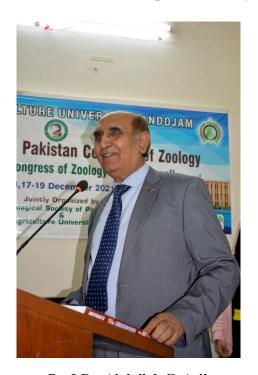
Participants of the congress



Participants of the congress



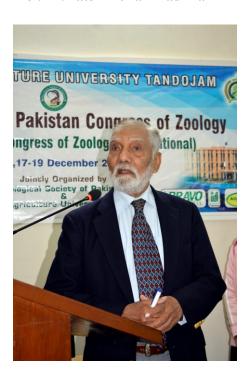
Prof. Dr. Muhammad Iqbal Chaudhary



Prof. Dr. Abdullah G. Arijo



Prof. Dr. Fateh Muhammad Marri



Mr. Abdul Aziz Khan



Prof. Dr. Muhammad Perwaiz Iqbal receiving Lift Time Achievement Award from Prof. Dr. Muhammad Iqbal Chaudhary



Prof. Dr. Juma Khan Kakar receiving Life Time Achievement Award from Dr. Muhammad Iqbal Chaudhary



Mr. Abdul Aziz Khan receiving souvenir from Dr. Muhammad Iqbal Chaudhary



Prof. Dr. Abdul Rehman receiving souvenir from Dr. Muhammad Iqbal Chaudhary



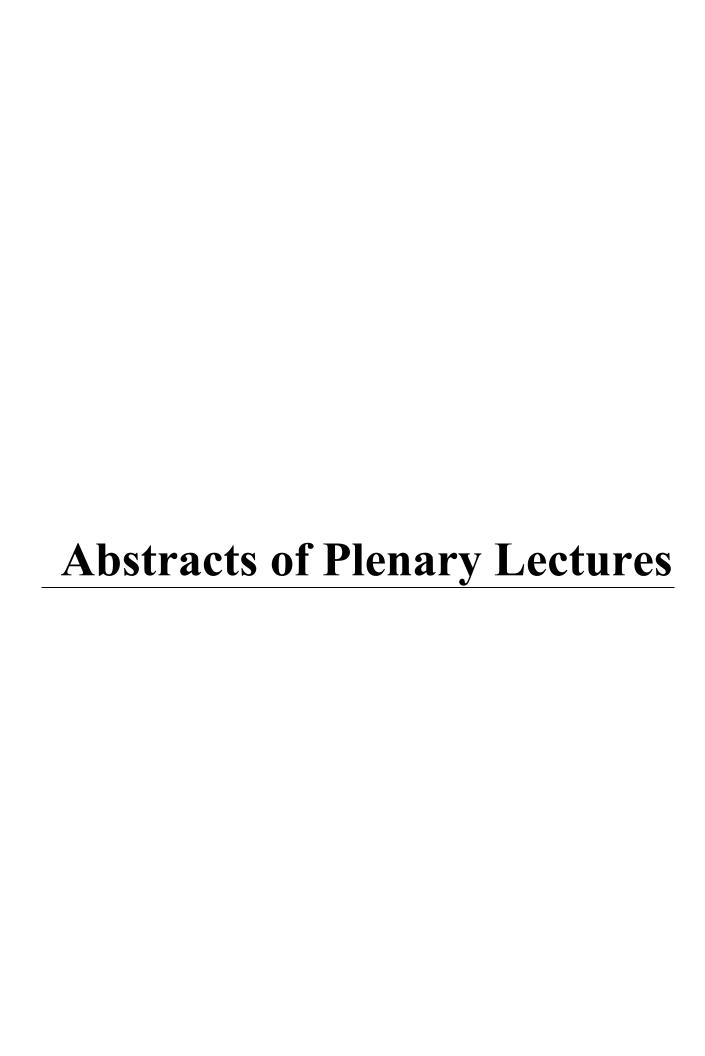
Dr. Mohammed Sajid Hamid Akash receiving Prof. Dr. A.R. Shakoori Gold Medal from Dr. Muhammad Iqbal Chaudhry



Prof. Dr. Abdullah G. Arijo receiving souvenir from Dr. Muhammad Iqbal Chaudhary



Dr. Riffat Sultan receiving a souvenir from Dr. Muhammad Iqbal Chaudhary





# DIVERSITY, SPECIATION MECHANISMS AND THE SPECIES CONCEPT IN *PARAMECIUM* (CILIOPHORA)

### Aleksei Potekhin

Department of Microbiology, Faculty of Biology, Saint Petersburg State University,
Saint Petersburg, Russia
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The species concept remains debated in modern zoology, and, in particular, in protistology. The overview on Paramecium as a model for species concept will be provided. Paramecium is one of the best studied genera of ciliates including fourteen valid morphological species. Most of these species are subdivided into reproductively isolated groups, the so called syngens. In last twenty years many Paramecium species have been re-described or discovered, including several new morphological and cryptic species. The hidden Paramecium diversity provides a perfect example that both morphological similarity or plasticity can be misleading in species identification, and molecular data are not always reliable by themselves. Ciliates serve as an remarkable example of the group where speciation is actively ongoing, and even within Paramecium there are several mechanisms driving initial speciation.



# FUNCTIONAL CHARACTERIZATION OF FATTY ACID DESATURASES (FADS2) IN COMMON CARP (CYPRINUS CARPIO): INSIGHTS INTO CATALYZING EFFICIENCY BASED ON IN VITRO, IN VIVO AND DOCKING SIMULATIONS

### Chenru Yang<sup>1,2</sup>, Shangqi Li<sup>1</sup>, Yaxin Wang<sup>1,2</sup>, Yujie Zhao<sup>1,2</sup> Senbiao Fang<sup>1</sup>, Jiongtang LI<sup>1</sup>, Yan Zhang<sup>1\*</sup>

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Long-chain polyunsaturated fatty acids (LC-PUFA), especially the n-3 polyunsaturated fatty acids, are essential compounds that important in numerous metabolic and physiological processes ensuring normal cellular function for vertebrate including fish. Common carp is cultured commercially worldwide, and it can be able to conversion dietary C18 precursor fatty acids to arachidonic acid (ARA), EPA, and DHA, likewise other carp species. cDNAs for two fads had been identified in common carp. We presented a comprehensive investigation of the function of common carp two fads genes, fads2a and fads2b combining docking simulation, in vivo, in vitro. The accurate of enzyme activity, conversion ratio, spatial express pattern, and promoter region activity were studied in this study. We identified  $\Delta 5/6$  activity for these two genes, fads2a and fads2b, combing predication of protein modeling and verification of recombination yeast expression system. Moreover, we estimate the conversion ratio of common carp fads2a and fads2b by overexpressing system in zebrafish. Taken together with the study of express pattern of these two genes, we explore the two genes molecular mechanisms.



### THE EFFECT OF EXTENDING LIGHT PERIOD IN FISH CULTURE

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In this century, It is essential that people are fed with the right protein sources for their health and for the continuation of their future generations. In this respect, as a source of animal protein, all aquaculture species, especially fish, have a significant importance. Nowadays, as the natural water resources are decreasing day by day and the need for animal protein is increasing due to the increasing population. In this point, aquaculture play an important role and It is a good alternative to meeting the need for fresh fish. The total world aquaculture is approximately 171 million tons (170,995,437 tons, excluding aquatic plants). Approximately 80 million tons (80.071.894 tons) and marine and inland water fish (51.368.288 tons) contribute to this amount through aquaculture. Aquaculture has been declared by FAO as the fastest growing food production sector in the world. It is estimated that fish production will continue to increase around 10% every year in the next 10 years. The share of aquaculture in Turkey's total production is in Turkey 43.83%. Trout (39,6%), sea bream (22,09%), sea bass (36,15%) and carp are the most cultivated species in our country. These species; suitable for successful aquaculture applications. In addition, production success is higher compared to other fish species. Therefore, more research is being done on these species. Studies are generally concentrated around feeding, disease and breeding studies. Some strategies are being developed in order to reach the market size of the fishes grown in the shortest time and to produce them at the lowest cost. For example, there are applications made by changing environmental conditions. In addition to changes in water quality such as temperature, salinity, pH, O2, adaptations to structural areas such as lattice or concrete pond have been investigated. The results of all these studies show that there are species-specific answers. Light has an effect according to the species and characteristics of living organisms on regulating the feed intake, migration time, gonadal stimulation, ovulation, growth, coloration and especially the respiration of aquatic organisms. Parameters such as temperature and nutrients, which include environmental factors, show more or less changes annually, while changes in natural light remain the same every year. Natural light changes regularly depending on the latitude, day, month and season. Natural or artificial light, which has a wide working area, has been the subject of great interest and research on various fish species cultivated by many researchers. Recent research has focused on observing response to lighting as stress, physiological status, behavior and growth performance of farm fish. In this presentation, the effects of artificial lighting and light time increase on the development and feed evaluation of some species will be examined.



# SOME INNOVATIVE TECHNOLOGIES IN AQUACULTURE WITH RESPECT TO FISH FEEDS AND CAGE FARMING

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Technological changes are changing the life of the people very quickly. Industrial changes and technological changes together are directing the way of production. In a World with smart Technologies and artificial intelligence, it is expected that many people will change their professions. People will mater on specific topics and will not work 40 hours in a week, only 30 hours will be enough for everything. In the same way aquaculture production volume is getting more and more in volume by both introducing new species, genetical improvements, and introducing new Technologies to fish farms and research environments, such as using innovative methods for production fish feeds or remote tracking and observing devices in a fully controlled farm. The relation of aquaculturally improvements to Industry 4.0 or Society 5.0 is due to its unique property being both social and science discipline. Since aquaculture deals with live material for human consumption. Some of the new technologies introduced directly or indirectly to aquaculture production will be highlighted for feed preparation, individual fish farming, and cage farming aspects.



# ZOONOTIC AND ZOOANTHROPONOTIC POTENTIAL OF SARS-COV-2

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Transmission of a pathogen from animal-to-human (zoonosis) and human-to-animal (zooanthroponosis) are currently leading threats for both veterinary and human public health. These threats can be testified by the fact that ~60% of emerging infectious diseases are of zoonotic origin and ~70% of these originate from wildlife. The importance of studying zoonotic and zooanthroponotic diseases is further highlighted by the emergence of multiple variants of SARS-CoV-2. A range of animal species have been verified for SARS-COV-2 and other zoonotic infections either in vitro or in vivo. However, molecular bases of such broad host spectrum of these infections remained elusive. In this lecture, I will layout key features of SARS-COV-2 that make the virus living example of zoonotic and zooanthroponotic pathogen.



### PARASITIC INFESTATION IN FANCY BIRDS: AN EPIDEMIOLOGICAL SURVEY IN PUNJAB

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There are different fancy birds i.e. pheasants, partridges, quails, ducks, Parrots, pigeons, geese and doves that are reared for an ornamental purpose as well as pet, sports, meat, recreational, teaching of taxidermy, biological and medical experimentations. They are domesticated all around the globe in the villages, towns and cities. Effective management, health care and nutrition are necessary for the welfare of these species. Birds could be parasitized by a wide variety of endo-parasites i.e. nematodes, trematodes, cestodes, acanthocephalans, and protozoa. Severe infections of these endo-parasites in birds significantly cause droopiness, loss of weight and diarrhea. These parasites consume nutrients and vitamins resulting the decreased feed utilization, intestinal obstruction and toxins production in the host. So, parasites are the main cause of serious health issues or even deaths that are newly kept in the captivity or adopted into new environment. Parasites were identified through the feces by simple laboratory methods. Prevalence of the doves positive for the coccidiosis zin Jhang was found (240/320) 75% and for Capillaria was (80/320) 25\% in Jhang. No Mixed infection (Eimeria spp. + Capillaria spp.) prevalence was identified in doves. EPG counted through McMaster slide for the Coccidia was 700, while the EPG for *Capillaria* was 850 in Jhang of dove birds. The incidence of the cockatiels birds positive for the coccidiosis in Layyah found was (120/170) 70.5% and for Capillaria was (30/120) 17.6% while, mixed infection (*Eimeria* spp. + *Capillaria* spp.) prevalence was (20/120) 11.7%. The incidence of budgerigars Layyah parrots observed was (80/90) 89% for coccidiosis and for Capillaria was (10/90) 11% and no mixed infection (Eimeria spp. + Capillaria spp.) was

reported. Prevalence of the cockatiel and budgerigar identified for the parasite diagnosis in Jhang; both showed positive for the coccidian only and the prevalence of coccidiosis in cockatiels was (210/210) 100% and in the budgerigars was (120/120) 100%. EPG counted through McMaster slide for the Coccidia was 750, while the EPG for Capillaria was 700 in both cases of cockatiels and budgerigars of Layyah and EPG counted through McMaster slide for the Coccidia was 750 in both cases of cockatiels and budgerigars of Jhang. EPG count for the coccidian in both species of parrots was same in the both districts that showed very weak burden of the parasites. The prevalence of the Pigeons bird positive for the coccidiosis in Muzaffargarh was found 70.7% and for Capillaria was 15.7% in Muzaffargarh and the mixed infection prevalence was 13.4%. EPG counted for the Coccidia in Muzaffargarh was 750, while the EPG for Capillaria in Muzaffargarh was 700 in pigeons. No parasite was identified in (400) quail species from the Layvah district. This study represented that Coccidia and Capillaria were the most prevalent parasites found in the pigeons, doves and parrots species (cockatiels and budgerigars) in all the studied districts, but no parasite was found in the quails of Layyah district. This study recommends for the proper management and care of these birds' species in the studied districts to get risk of these parasitic infections to prevail these towards the other livestock populations and humans.



### HISTORICAL SWARMING OF LOCUST IN PAKISTAN: CAUSES AND CONTROL STRATEGIES

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Recent plague hit Pakistan for the first time in more than two decades and cause million rupees damage to economy. It is considered as big historical disaster to the agriculture and livestock. They are exceptional in mobility that's why a swarm of desert locust crossed Balochistan's desert area of Cholistan, Rahim Yar Khan, Bahawalpur, Thar Desert, Mirpurkhas, Umerkot, Nara Desert, Khairpur and Sukkur to the Indian border in single flight. Locusts remain a major food security challenge today throughout the world. Locusts, which caused a significant damage to Pakistan's agriculture back in the 1950s, 1960s and 1990s have once again swarmed a huge areas in Pakistan actually unusual, rains throughout Pakistan provided them a favourable environment, which aggravated this situation. Swarms of locusts are landing on vast fields of cotton, wheat, sugarcane and other standing crops in fertile parts of lower Sindh and Balochistan. It is estimated that million acres fertile land was badly affected by this sudden eruption although, earlier plagues persist for shorter time periods but recent plagues is largest one started from May 2019 and continued till November 2019. The desert locust eats a wide variety of crops and other plants, including a broad assortment vegetable and cereal crops, banana, citrus, groundnuts, fruit trees, and many others. Due to its vast reach and significance to agriculture, this species is often considered the most dangerous migratory pest in the world. Schistocerca gregaria is multivoltine, with up to 3 generations per season under favorable conditions. Typically, it takes several years for outbreaks to develop into a plague, but plagues can subside within 6 months. Locusts can travel 150 km in a day and typically migrate between seasonal breeding areas. Heavy rains can allow for population buildup within the recession zone. As vegetation rescinds, locusts aggregate,

which can lead to gregarization. But no detail study was available on exact causes of swarm and its managements from this region yet. The purpose of this study was: 1. Investigation on origin of swarming 2. Breeding zones of S. gregaria in Pakistan along with its occurrence and seasonal distribution 3. Examination of environmental factors that affect phase change, along with the wider impact of land use and management strategies that may unwittingly create environments conducive to outbreaks 4. Estimation of crops losses and its negative impact on food security in the region. Beside this, reproductive activities viz: copulation, oviposition, hatching, nymphal development, fecundity, fertility in solitarious and gregarious phases was also compared. I examine that solitarious phase locusts tend to be inactive and avoid each other, while gregarious phase individuals are more active and typically orient toward one another and consume the equivalent of their body weight (2 g (0.07 oz)) each day in green vegetation. Mostly the aerial operation was undertaken against this attack in Balochistan and some parts of Sindh but unfortunately situation in many areas is still depraved and out of control. Present study recommends that monitoring of Desert locust will be made possible through remote sensing, GPS, GIS, and Satellite data. Further, well-coordinated efforts such as: cultivation of nitrogen low plantation, cultural and physical operation and utilization of biocontrol agents against locust at regular bases are needed in order to avoid the production of swarming in future.



### TICK CONTROL: CONVENTIONAL STRATEGIES AND NOVEL APPROACHES

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Ticks, both soft and hard are the utmost important ectoparasites of livestock in tropical and sub-tropical countries of the world. They are blamed for severe financial losses both through the direct effects as blood-sucking and indirectly as vectors of diversified pathogens and toxins. Worldwide losses due to disease s transmitted by ticks and the costs of tick control have been estimated to be in the range of several billion annually. The importance of ticks is principally due to the ability to transmit a wide spectrum of pathogenic micro-organisms, such as protozoa, rickettsiae, spirochetes and viruses. In Pakistan, tick-borne protozoan disease s (e.g. Theileriosis and Babesiosis and Anaplasmosis) are the main health and management problems of domestic ruminants. Various control methods have certain limitations and in certain cases, the result on tick control are not encouraging. Therefore, local remedies, such as the use of ethnobotanical products are considered as suitable and safe replacement of tick control. This presentation is aimed at reviewing various methodologies with focus on the potential of local remedies towards tick control.



### MICROBACTERIUM SP. STRAIN 1S1 RESISTANCE STRATEGIES AGAINST METALS: A FEASIBLE APPROACH FOR ENVIRONMENTAL CLEAN-UP

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Microbacterium sp. strain 1S1, tolerated arsenite and arsenate upto 75 and 520 mM, was isolated from industrial wastewater. Metal uptake and surface adsorption into the bacterial cells challenged with 15 mM arsenite were confirmed by SEM, EDX, and FTIR analyses. Reduced and oxidized glutathione ratio, non-protein thiols and catalase were increased upto 40.0, 78.50, and 240 % under metal exposure. The bacterial cells were able to oxidize 98% arsenite after 96 h of incubation at lab scale while inactivated biomass removed 99%/10 h arsenite from the medium. The genes for arsenic oxidation aioB (arsenite oxidase smaller subunit) along with MoeB (molybdopterin biosynthesis protein) and erpA gene (Fe-S cluster insertion protein) were found on the chromosomal DNA of the strain. The 5 ArsC genes, one Acr3, 2 thioredoxin reductase (txrA, txrB), 3 arsenic regulatory genes (arsR1, arsR2), 4 phosphate transporting genes (psts, pstC, pstA, pstB), and one arsB gene which encode arsenite efflux pump were found on chromosomal DNA indicating that resistance could be ascribed to As-efflux system. MS analysis revealed that 24 proteins expressed while 42 proteins were suppressed when cells were challenged with 15 mM arsenite. Presence of multiple resistance strategies and highest arsenic oxidizing potential make strain 1S1 an impending foundation for green chemistry to exterminate environmental arsenite.

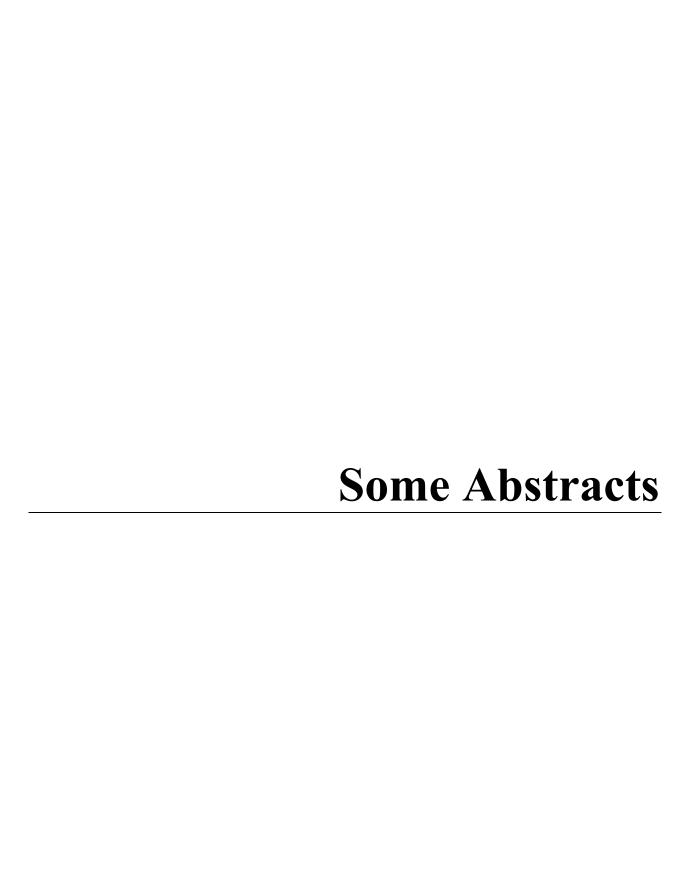


# EPITHELIAL – MESENCHYMAL TRANSITION: EFFECT OF METFORMIN CHLORIDE

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Glucose starvation is long being studied for its potential to control cancer metastasis. A decade ago the drugs interfering the glucose metabolism started gaining attention for their anticancer properties and metformin is one of them. Metformin chloride is a common antidiabetic drug. Different studies have used different concentrations on a variety of cancer cells. Our study mainly focuses on its effect on epithelial to mesenchymal transition gene markers and inducers in various cancer cell lines. For that purpose, different nontoxic concentrations (0.01 M, 0.05 M) to intermediate (0.1 M) and toxic concentrations (0.2 M, 0.5M) of this drug were prepared to be used against different cancer cell lines including SF767, HCT-116, MDA-MB231, and MCF7 along with normal Human Umbilical Cord Stem cells as control. In our study through various biochemical assays and gene expression studies, we found that the usual lower concentrations are not only less effective in an in-vitro model compared to higher dosages but can also increase cancer proliferation and upregulate the metastatic markers by activating STAT3 signaling rather than inhibiting it.



### **SECTION - I**

# CELL BIOLOGY, MOLECULAR BIOLOGY, GENETICS, PHYSIOLOGY, TOXICOLOGY, VIROLOGY

- 1. HERBAL MEDICINE, BIOCHEMISTRY, BIOTECHNOLOGY AND BIOINFORMATICS
- 2. CELL AND MOLECULAR BIOLOGY, CELL BIOLOGY, GENETICS
- 3. HUMAN AND ANIMAL DISEASES
- 4. MICROBIOLOGY
- **5. MOLECULAR BIOLOGY**
- **6. PHYSIOLOGY**
- 7. TOXICOLOGY
- 8. VIROLOGY

### 1. HERBAL MEDICINE BIOCHEMISTRY, BIOTECHNOLOGY AND BIOINFORMATICS

### STUDIES ON SCREENING OF SOLANUM NIGRUM FOR NUTRITIONAL, ANTIOXIDANT AND ANTIMICROBIAL ACTIVITY

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The study was designed to screen proximate analysis and biological activity of whole plant of *Solanum nigrum*. Proximate analysis of plant was performed to evaluate the nutritional values of *Solanum nigrum* which were 8.1, 24.9, 14.8, 37 and 28.6 for moisture, ash, protein, fat and fiber and plant showed highest concentration of fiber 37% and lowest concentration of moisture 8.1%. It was tested for antioxidant activity using 2, 2- diphenyl-1-picrylhydrazyl (DPPH) assay method. The antibacterial activity was detected by agar well diffusion method and highest values were obtained for *Bacillus spizizenii*, *Pseudomonas aeruginosa*, *Staphylococcus aureus* and *Escherichia coli i.e* 14.3±0.58, 11.3±0.58, 10.6±0.58 and9.3±0.58 respectively. Overall results revealed that *solanum nigrum* exhibits excellent nutritive value along with potent antioxidant and antimicrobial activity. It can be used in medicine as antimicrobial as well as in food supplements to prevent their oxidation and rancidity of food.

## EFFECTS OF TRIGONELLA FOENUM-GRAECUM SEEDS ON LIPID PROFILE IN OBESE FEMALES

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To reduce the lipid profile in obese females by the Fenugreek seeds. This is one month follow up study from 4th May to 5th June 2019. Prior two days to start Ramadan draw blood samples after 8 hours fasting for lipid profile estimation from 50 obese female subjects. Anthropometric measurements obtained from blood sample donors include height, weight, arm, hip, thigh, chest and waist circumference and also recorded blood pressure. Same protocol was used at the last day of Ramadan to compare pre and post fenugreek effect on selected subjects. All measurements were taken before and after using fenugreek for about a month of Ramadan. A paired t-test was run to analyze the mean differences before and after using [fenugreek]. The means decreased for all variables, except HDL cholesterol, which is the good cholesterol in the body and is expected to increase with an improvement in overall health. This indicated that fenugreek worked and, on average, all study participants improved their body measurement. Measurements taken before and after using fenugreek were strongly and positively correlated with each other (r > 0.89 for all comparisons, p < 0.0001). The difference between the means before and after using fenugreek was significantly different than 0 for all variables except cholesterol. For example, the mean difference between weight before and after using fenugreek was 2.349 kg (t42 = 7.246, p < 0.0001). In other words, other than cholesterol, all measurements dropped significantly after using fenugreek and we would reject the null hypothesis for all except cholesterol. Fenugreek seed give a positive result on human health for reduction in over-weight. The fenugreek refer as best herbal remedy because its having no after effect on human health and also it is very cheaply available in markets.

### EFFECT OF REPLACEMENT OF SUNFLOWER MEAL WITH MORINGA OLEIFERA LEAF POWDER ON IMMUNITY AND GUT MICROFLORA OF BROILERS

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The present study was conducted to evaluate the effect of replacement of sunflower meal with *Moringa oleifera* leaf powder on immune system and gut microflora of broiler chickens. The experiment was conducted on one hundred day old broiler chicks. The chicks were distributed into four treatment and one controlled group with 20 birds in each group. Groups A, B, C, D, and E were offered 5%, 3.75%, 2.5%, 1.25% and 0% of *Moringa oleifera* leaf powder per kg of feed respectively. The experimental groups were observed for the performance parameters, antibody titer against Newcastle disease (ND) and Avian Influenza (AI), relative weight of lymphoid organs (bursa, spleen and thymus) carbon clearance assay, cutaneous sensitivity response to avian tuberculin and quantification of gut microflora. Results showed that *Moringa oleifera* leaf powder affected the feed intake, final body weight, carcass weight and relative weight of lymphoid organs. *Moringa oleifera* leaf powder supplementation also modulated the immunological parameters like carbon clearance assay, cutaneous sensitivity response to avian tuberculin and antibody titers against ND and AI, as well as altered the gut microflora of broilers. In conclusion, *Moringa oleifera* leaf powder had positive impact on performance, gut microbiota and immune responses of broilers.

# IN VIVO HAEMATOPROTECTIVE AND ANTI-INFLAMMATORY EFFECTS OF CORDIA OBLIQUA EXTRACT

### Shafaat Yar Khan\*, Mas'ab Umair Rana, Muhammad Khalid Mukhtar, Sajida Batool, Samra Ashraf Rana, Aleem Ahmad, Sadia Azam, Kafeel Qasim and Breha Kazmi

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During present study, haematoprotective and anti-inflammatory properties of methanolic leaves extract of Cordia obliqua (Lasora) were evaluated in mice. Chromium (Cr) induced haematotoxicity was used to assess haematoprotective potential whereas hioglycolate-induced peritonitis model was used to evaluate antiinflammatory activity. Haematotoxicity was induced using Cr (20 mg/kg) through intraperitoneal inoculation while oral route was used to administer methanolic leaves extract of C. obliqua (50 mg/kg) in mice during the experiment of two weeks. Hematological parameters including TEC, Hb, HCT, MCV, MCH, MCHC, and DLC were evaluated in the animals exposed to Cr with or without leaves extract of C. obliqua. Cr treatment led to a significant reduction of TEC, Hb, HCT, MCV, MCH, MCHC, TLC, lymphocytes and monocytes while leaves extract of C. obliqua along with Cr prevented this reduction significantly. Cr treatment exhibited a trend of increase in TLC and neutrophils while methanolic leaves extract of C. obliqua along with Cr prevented this increase in TLC and neutrophils. Thioglycollate treatment induced an increase in leukocytes infiltration and IL-6 level in peritoneal lavage of mice in hioglycolate-induced peritonitis model while leaves extract of C. obliqua (50 mg/kg) along with hioglycolate significantly prevented this increase in leukocyte infiltration and IL-6 level in peritoneum exhibiting its strong antiinflammatory potential. Results revealed that methanolic leaves extract of C. obliqua have effective haematoprotective activity against Cr induced haematotoxicity and possess a strong anti-inflammatory potential in hioglycolate induced peritonitis model in vivo.

### SUB-LETHAL EFFECTS OF INSECTICIDES ON ESTERASES OF SPIDER GEA SPINIPES (ARANEAE: ARANEIDAE)

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Spiders as general predators are well characterized that particularly feed on crop pest within the agroecosystems and provide indirect benefits to humans by pest consumption. Field application of non-natural insecticides minimized the abundance of insect pests but also drastically effects the spider growth and could lead to developed resistance in pests. Gea spinipes C. L. Koch, 1843 is an orb-weaver spider with web hardly 1 foot from the ground around grasses and low shrubs. For this study, live specimens were collected from the field areas of University of The Punjab, Lahore. Five different insecticides were used with different doses and concentration of active ingredient to estimate the level of detoxifying enzymes such as Acetylcholine (AChE) and Carboxylesterase (CarE). For this purpose 0.5 µl drop of each dose was applied on the thorax of spider and left for 12 hours and droplet of water for control group. The reduced level of AChE as compared to control group was estimated by use of insecticides such as fipronil, cypermethrin, spinetoram and thiamethoxam while the level of CarE was elevated as compared to control group with same insecticides. Spiromesifen showed different results in which the level of both AChE and CarE was reduced as compared to their control group. Doses of 1% and 0.1% of all insecticides caused mortality in few spiders prior to experiment while doses of 0.01% and 0.001% showed no mortality and they are subjected as sub-lethal doses. These results suggest that lethal or sub-lethal doses of insecticides drastically effect the population of natural predator Gea spinipes. Insecticides that have sub-lethal effects on spider can reduce their predatory potential depending on type of insecticides and predator species.

# MULTIPLE HEAVY METAL RESISTANT MICROCOCCUS LUTEUS STRAIN AS2 ISOLATED FROM INDUSTRIAL WASTE WATER AND ITS POTENTIAL USE IN ARSENIC BIOREMEDIATION

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Multiple heavy metal resistant bacterium, *Micrococcus luteus* strain AS2, was isolated from industrial waste water of District Sheikhupura, Pakistan. The isolated bacterium showed MIC of 55 and 275 mM against arsenite and arsenate. The bacterial strain also showed resistance against other heavy metal ions i.e. lead, cadmium, chromium, mercury, nickel and zinc apart from arsenic. The optimum temperature and pH was 37°C and 7, respectively. The antioxidant enzymes such as catalase were significantly increased under arsenite stress. The increased in 43.9% of GSH/GSSG and 72.72% of non-protein thiol was determined under15 mM arsenite stress. Bacterial genome was sequenced through illumina and nanopore and genes related to arsenic and other heavy metals were identified and blast (tblastx) on NCBI. Through scanning electron microscope, no morphological changes were observed in bacterial cells under arsenite stress. The peaks appeared in EDX showed that there is surface adsorption of arsenite in bacterial cell while it was confirmed from FTIR-analysis that there is some interaction between arsenite and functional groups present on the surface of bacterial cell. The SDS-PAGE analysis of whole cell proteins under 15 mM arsenite stress clearly revealed that there is up-regulation of some proteins in ranged of 60 to 34 kDa. The bioremediation efficiency (E) of bacterial biomass was 72% after 2 h and 99% after 10 h. The bioremediation efficiency of bacterial biomass is an indicator for the isolated bacterium to employ as a potential candidate for the amelioration of sites contaminated with arsenic.

### DIETARY SELENIUM YEAST SUPPLEMENTATION ALLEVIATES HIGH GRAIN DIET TRIGGERED OXIDATIVE STRESS AND INFLAMMATORY RESPONSE IN THE CECUM OF GOAT

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The current research was designed to estimate the influence of selenium (Se) on mechanism of high grain (HG) diet induced adverse effects in the cecal epithelium of goats. Eighteen cross-bred goats, randomly allocated into three groups (n=6/group) either fed low grain (LG, roughage: concentrate ratio 65:35), high grain (HG, 35:65) or HG supplemented with Se (HGSe) diets for 10 weeks. Se was added at the rate of 0.5mg.kg<sup>-1</sup> diet. The concentrations of individual and thus total short chain fatty acids (SCFA) was significantly increased (P < 0.05) with simultaneous decrease in pH of cecal fluid in HG and HCSe compared with LG. The criteria of epithelial injury showed loss of epithelial integrity, inflammatory cell infiltration and loss of goblet cells in cecal mucosa associated with higher lipopolysaccharide (LPS) concentrations in cecal fluid and plasma of HG diet group, whereas the addition of Se in HG diet alleviated such damages. Compared with LG, the HG diet elevated malondialdehyde (MDA) level with concurrent decrease in glutathione peroxidase (GSH-Px) and superoxide dismutase (SOD) activities suggesting epithelial injury leading to oxidative stress while, Se supplementation attenuated these changes and improved antioxidant status in colonic epithelium of HGSe. Compared with LG, the HG diet induced inflammation by modulating gene expression of proinflammatory interleukins (ILs) and cytokines; whereas, Se supplemented HG diet alleviated such changes by elevating the expression level of anti-inflammatory genes in the colon of goats fed HGSe diet. This data demonstrated that addition of Se in HG diet reduced mucosal damages via showing antioxidant and anti-inflammatory effect in cecal epithelium of goat.

# ASSESSMENT OF CHOLINESTERASE ACTIVITY OF BIRDS INHABITING PESTICIDE EXPOSED CROPLANDS OF GUJRAT, PUNJAB, PAKISTAN

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The study was undertaken to explore seasonal distribution of five bird species i.e Black drongo, Common babbler, Jungle babbler, White wagtail and Red vented bulbul; and to assess the activity level of brain cholinesterase (ChE) in bird species inhabiting agricultural landscapes of Gujrat having the history of pesticide applications. Data on the diversity of bird species were collected along 1km line transects during 2017-18 on monthly basis. Spectrophotometric method for determination cholinesterase (ChE) activity in brain tissues of the birds was used to assess the impact. The distribution pattern showed higher level of abundance of all bird species in the winter season as compared to the summer at the cropland. Bird count showed a total of 1302 individual with dominance (0.788), Shannon-Weaver diversity index (1.579), and evenness (0.97). The bird species counts showed that black drongo (26.26%) followed by common babbler (23.19%) and jungle babbler (21.42%). The lowest relative abundance was shown by white wagtail (12.74 %). The change of cholinesterase (ChE) activity in the brain of birds present at the croplands of Gujrat was compared with the bird species inhabiting at Deva Vatala National Park (DVNP). These bird species were shot dead, identified, dissected to collect the brain; and stored for the spectrophotometric analysis. Brain cholinesterase activities of bird species in the ascending order were recorded i.e. jungle babbler (27.6/µmol/min/g), white wagtail (47.9/µmol/min/g), babbler  $(54.8/\mu mol/min/g)$ , red vented bulbul (55.9/µmol/min/g) and black (79.9/µmol/min/g) whereas a different order of brain cholinesterase activity was observed in the samples

collected from species inhabiting DVNP. The order of activity level of ChE inhibition is the highest in jungle babbler (53%) followed by common babbler (35%), red vented bulbul (18%), white wagtail (15%) and black drongo (7%). The higher activity level of cholinesterase was observed in black drongo whereas the lowest was recorded in jungle babbler captured from the croplands of Gujrat. The brain cholinesterase inhibition under different protected ecosystems and agricultural landscapes suggest environmental contamination and its impact on avifauna diversity. The study also emphasizes on the presence of pesticide free zones to protect the bird's biodiversity.

### COMPARATIVE ANALYSIS OF CATALASE AND PEROXIDASE ACTIVITY IN DIFFERENT TISSUES OF CYPRINUS CARPIO FROM RIVER CHENAB

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Present research work was carried out on *Cyprinus carpio* to evaluate pollution induced oxidative stress by using stress markers catalase and peroxidase. Fish samples were collected from selected sites of River Chenab and semi-intensive pond of Fisheries Research Farms, University of Agriculture, Faisalabad (UAF) as a reference site. Various physico-chemical parameters viz., temperature, electrical conductivity, dissolved oxygen and pH of water were recorded on the spot. Selected organs such as liver, gills, heart, muscle and kidney were extracted from the fish at the sampling site. The extracted organs were stored in crushed ice box after placing them in tagged zipper bags and brought to Aquaculture Biotechnology Lab, UAF for further analysis. The activity of catalase and peroxidase was measured on spectrophotometer at 240nm and 470nm, respectively. Data was analyzed by using ANOVA and Tuky's HSD post Hoc test by using Statistix 8.1 software. Highly significant variations were observed in the activity of both enzymes at p<0.05. The activity of both enzymes was observed in the following order liver>gills>kidney>muscle>heart for catalase and liver>gills>kidney>heart>muscle for peroxidase. The activity of catalase was found significantly higher in liver tissues (287.62±.32UmL-1) than peroxidase (210.8±0.06 UmL-1) at Trimmu Barrage as compared to other sites. The inferences of present study will be helpful to understand how stress markers can be used to evaluate the influence of pollution in aquatic environment.

# OVERPRODUCTION AND CHARACTERIZATION OF RECOMBINANT GroEL AND GroES FROM GEOBACILLUS THERMOPAKISTANIENSIS TO DEVELOP A PROTEIN REFOLDING SYSTEM

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Homologues of bacterial of GroEL and GroES genes encoding GroELGt (ESU72018) and GroESGt (ESU72017), from Geobacillus thermopakistaniensis were cloned and expressed in Escherichia coli. The purified proteins possessed the ATPase activity and were able to refold the denatured insoluble aggregates of α-amylase from Bacillus licheniformis into soluble and active form. GroELGt and GroESGt were found to successfully enhance the thermostability of commercial porcine heart malate dehydrogenase. Recombinant E. coli cells expressing GroELGt gene enhanced their thermotolerance. Recombinant alcohol dehydrogenase from Bacillus subtilis strain R5 in E. coli, was produced as soluble and active by co-expressing with GroELGt by using pETDuet-1 vector system, which otherwise produced as insoluble aggregates. GroELGt found to assist the folding of nascent protein in E. coli with the help of host co chaperonin. This system can be used for soluble production of recombinant proteins during expression which otherwise are produced in insoluble form in E. coli. To the best of our knowledge this is the first report on functional characterization an applications of chaperonins from genus Geobacillus.

### SESQUITERPENE LACTONES AS NOVEL STAT3 INHIBITORS

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Signal transducer and activator of transcription 3 (STAT3) is a transcriptional factor which is implicated in a wide range of cellular functions such as cell proliferation, survival, differentiation, metabolism, and immunity. Unlike normal cells where activation of STAT3 is strictly regulated, in cancer cells, it is constitutively activated and plays vital role in tumor induction, progression and drug resistance. Compelling evidence from a plethora of research reports has validated STAT3 as an attractive cellular target for anticancer drug discovery. To date, several STAT3 inhibitors have been discovered and characterized, however; none of them has been approved by FDA for clinical use in the treatment of cancer, indicating that mere inhibition of STAT3 is not sufficient enough to effectively set the cancer cells on the road to ruin. Therefore, exploring novel STAT3 inhibitors with potential anticancer activity are highly desirable. Sesquiterpene lactones are plant-derived natural bioactive molecules with a range of biological and pharmacological activities including anti-bacterial, anti-fungal, anti-inflammatory and anti-cancer activities. The presence one or more than one alkylating centres has made this class of compound as an attractive and promising leads for drug development. Here, in the present study, we found that sesquiterpene lactones are potent STAT3 inhibitors. The STAT3 inhibitory activity of sesquiterpene lactones was detected by molecular docking, Western blot and SPR analyses. Collective data indicated that sesquiterpene lactone could selectively bind with STAT3 and thereby inhibits STAT3 activation

# BIOACCUMULATION OF TRACE AND TOXIC METALS FOUND IN DIFFERENT ORGANS OF TWO CARP FISH SPECIES FOUNDS IN RIVER INDUS AT GUDDU BARRAGE KASHMORE

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The aim of study was to evaluate concentrations of heavy metals in fish samples from Indus River at Guddu Barrage District Kashmore. Selected metals Iron Fe, Zinc Zn, Copper Cu, Nickel Ni, Cobalt Co, Chromium Cr, Cadmium Cd and Lead Pb were determined in Gills, Liver and Muscles of freshwater fish species Catla catla, Labeo rohita found in Indus Guddu Barrage. Fish sample were caught by professional fishermen by using different size of fish nets. The samples of both species of five different weights were collected i.e. 200g, 400g, 600g, 800g, and 1000g. The samples were stored in an Ice box (at 4°C) and were transported to the laboratory for dissection and dried and homogenous digestion methods were carried out for elements viz. Iron Fe, Zinc Zn, Copper Cu, Nickel Ni, Cobalt Co, Chromium Cr, Cadmium Cd and Lead Pb concentration. Analysis of samples was performed by using Atomic Absorption Spectrometry, Perkin Elmer AA-800 in High Tech: Institute of Chemistry, Shah Abdul Latif University, Khairpur, Sindh, Pakistan. Findings of study indicates that order of concentration of elements such as Copper, Nickel, Cobalt, Chromium, and Cadmium were found high in Labeo rohita> Catla catla fish organs. While order of concentration of lead was noted high in Catla catla > Labeo rohita organs. In case of Iron the order of concentration was observed high in Labeo rohita > Catla catla fish samples. Order of concentration of Zinc was found maximum in Catla Catla> Labeo rohita fish samples. Concentration of elements were observed maximum in samples digested by Homogenous method as compared to dried digestion method may be due differ in acids combination. Concentration of all trace and toxic elements were found within the recommended daily allowance (RDA) Guide line as proposed by WHO/FAO.

### ASSESSMENT OF OVARIAN OXIDATIVE STRESS AND HEPATOTOXICITY INDUCED BY DEPOT-MEDROXYPROGESTERONE ACETATE AND PHARMACOLOGICAL INTERVENTION OF VITAMIN A AND C IN RABBITS

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This study was conducted to examine the ameliorating potency of combined vitamin A and C against depot-medroxyprogesterone acetate (DMPA) induces ovarian oxidative stress and Hepatotoxicity in rabbits. DMPA give rise to free radical injury and oxidative stress in different tissues which is cured by free radical scavenger and antioxidants. Combines Vitamin A and C are classified as natural antioxidants. Therefore, to attenuate the effects of DMPA are treated by combined vitamin A and C. Twelve female rabbits having age 5-6 month and weighing 3-4 kg were divided into four groups (n=3 rabbits in each): Control (I) (untreated); depot-medroxyprogesterone acetate (DMPA) (II), 50 mg/ per rabbit/week; Vitamin A+C (III), (Vit. A= 1000 IU /day/rabbit) (Vit. C= 10 mg/day) depot-medroxyprogesterone acetate (DMPA), 50 mg/ per rabbit/week; Vitamin A+C, (Vit. A= 1000 IU /day/rabbit) (Vit. C= 10 mg/day) (IV). The treatment was performed for five weeks to examine, ALT, AST and ALP for liver damage; SOD and MDA as a oxidative stress, and Leptin, Adiponectin and body weight as obesity marker. Our result reveled that DMPA enhanced the ALT, AST, ALP, MDA, Leptin, and body weight, while treatment of combined Vitamin A and C significantly (p <0.05) controlled this enhancement. The SOD and adiponectin level was insignificantly decreased when compared with Control group. This decreased SOD and Adiponectin was significantly prevented by combined Vitamin A and C.

# FABRICATION OF POLY (BUTADIENE-BLOCK-ETHYLENE OXIDE) BASED AMPHIPHILIC POLYMERSOMES: AN APPROACH FOR IMPROVED ORAL PHARMACOKINETICS OF SORAFENIB

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Sorafenib (SFN), a hydrophobic anticancer drug, has several limitations predominantly poor aqueous solubility and hepatic first-pass effect, limiting its oral delivery that results into several other complications. Present study aims to develop Sorafenib loaded polymersomes using Poly butadiene block poly ethylene oxide (PB-b-PEO), an amphiphilic co-block polymer. Prior to drug loading, critical aggregate concentration (CAC) of 'polymer was calculated for stable formulation synthesis. The developed SFN loaded PB-b-PEO polymersomes (SFN-PB-b-PEO, test formulation) characterized by DLS and cryo-TEM showed particle size  $282.88\pm22.61$  nm, polydispersity (PDI) of less than  $0.29\pm0.08$  and membrane thickness of about 20 nm. SFN-PB-b-PEO demonstrated encapsulation efficiency of  $71.42\pm11.98\%$ . SFN-PB-b-PEO polymersomes showed sustained drug release up to 144 h. Formulation remained stable for 3 months in suspension form. *In vitro* cytotoxicity against HepG2 cells showed 1.7 folds improved toxicity compared to SFN suspension. In addition, oral administration of SFN-PB-b-PEO in BALB/c mice showed increased  $C_{max}$  and  $AUC_{0-96}$  by 1.7 and 2.77-fold respectively (p<0.05) compared to those of SFN suspension (reference formulation). Findings suggest that the SFN-PB-b-PEO polymersomes can be a potential candidate for oral delivery of SFN.

### PRODUCTION AND PURIFICATION OF A PULLULANASE ENZYME FROM BACILLUS THURINGIENSIS

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In the present study production and purification of an extracellular pullulanase enzyme from *Bacillus thuringiensis* was carried out for use in various industrial processes. To get the maximum production of pullulanase enzyme, culture conditions like, pH of medium, incubation temperature, time of incubation, media composition and size of inoculum for *B. thuringiensis* were optimized. Maximum production of pullulanase enzyme (5.71 U/ml/min) was obtained in the medium containing tryptone as carbon and energy source having pH 6.0 when it was inoculated with 3% overnight grown inoculum and incubated at 37°C for 24 hours. Optimal conditions for pullulanase enzyme activity were also determined and maximum enzyme activity (8.584 U/ml/min) was found with 4% pullulan as a substrate in phosphate buffer of pH 7.0 at 50°C after 20 minutes of incubation. Purification of pullulanase enzyme was achieved to homogeneity by ammonium sulphate precipitation as well as by ion exchange chromatography and a distinct band of 97 kDa was analysed by SDS-PAGE. Purification fold of purified enzyme was calculated as 16.83 with 37% yield and 45.45 U/mg specific activity. These results are useful for large scale production of pullulanase enzyme from *Bacillus thuringiensis* for use in industrial applications.

### LACCASE PRODUCTION BY LOCALLY ISOLATED ASPERGILLUS NIGER AND ASPERGILLUS ORYZAE UTILIZING AGRO WASTES

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In this study laccase enzyme production was carried out from locally isolated Aspergillus species by solid state fermentation utilizing agriculture wastes as substrates for use in various industrial processes. Two Aspergillus strains, *Aspergillus niger* and *Aspergillus oryzae* were isolated from various food samples and identified by morphological and molecular means. To get the maximum production of laccase enzyme from both strains, various production parameters were optimized like pH of the growth medium, temperature of incubation, time of incubation of culture, inoculum size and substrate composition. Maximum production of laccase enzyme (7.45 U/ml/min) from *A. niger* was achieved in wheat bran medium at 28°C after 72 hours of incubation when pH of the medium was set at 5.0 and inoculation was carried out with 5% spore suspension. While maximum production of laccase enzyme (15.241 U/ml/min) was achieved by *A. oryzae* also in wheat bran medium at 28°C after 72 hours of incubation when the pH of the medium was set at 7.0 with 5% inoculum size. Optimal enzyme conditions were pH 7.0, temperature 30°C and time of incubation 30 minutes for *A. niger* and pH 6.0, temperature 40°C and time of incubation 30 minutes for *A. oryzae*, respectively. These results showed the potential of *A. niger* and *A. oryzae* for large scale laccase production for use in various industrial applications.

## ANALYSIS AND EVALUATION OF CARDIAC ENZYME PROFILE AND SERUM LIPID PROFILE IN PATIENTS TO FIND OUT ACUTE CORONARY SYNDROMES (ACS)

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Chest pain is a non-specific grievance and is the most common reason for patients looking for urgent medical care. The present study is to find out the prevalence of symptoms and disease situation with which patients visit the

outpatient department of the largest tertiary care hospital in Punjab, Mayo Hospital, Lahore. Samples of 207 patients having chest pain & lipid profile were collected and analyzed to check out the biomarker level of all cardiac and lipid enzymes. Different parameters like AST, LDH, and V.L.D.H were analyzed. Descriptive analysis of prevalence result was done on socio-demographic variables on statistical software SPSS 21. Other statistical test including unstandardized beta, standard error test, T test, Adjusted R square and F test values were also performed along with its significance showing goodness of fit of model. Results showed that correlation between LDH and V.L.D.H was negative and between LDH and CPK was strongly positive and significant. Conclude that when patient do more exercise their cardiac problem reduced.

# METABOLIC PROFILING OF SECONDARY METABOLITES PRODUCED BY *BACILLUS* SPP. EXHIBITING PLANT GROWTH PROMOTING AND BROAD-SPECTRUM ANTIMICROBIAL ACTIVITIES

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Phyto-microbiomes have been well explored in providing wide range of beneficial services to the plants leading to the enhancement of the plant growth. Among these rhizomicrobiome communities, the best understood and characterized are Bacillus spp. which through a variety of mechanisms directly and indirectly aid in plant nutrition. Keeping in view the multifaceted beneficial traits exerted by Bacillus spp., this study targeted the detailed polyphasic characterization of three root-colonizing Bacillus strains. Based on 16S rRNA sequencing, isolate SB-1 (sugarcane endophyte) was characterized as B. amyloliquefaciens, A-2 (rice rhizosphere) as B. subtilis, and A-3 (corn rhizosphere) as B. tequilensis. The strains exhibited broad-spectrum antifungal abilities and inhibited 43-86% growth of eight fungal pathogens tested *in-vitro*. All strains produced IAA in the range of 0.067-0.147 μM and were positive for the production of extracellular enzymes including protease, cellulase, and lipase. Moreover, B. amyloliquefaciens SB-1 solubilized 23.2±0.21 µg/mL of tri-calcium phosphate and ZnCO<sub>3</sub> (SI=2.1±0.18). UPLC-ESI-MS/MS analysis revealed the production of surfactins, iturins, fengycins, macrolactins and bacillomycin-D, which were further confirmed by amplifying the genes involved in the biosynthesis of these antimicrobial lipopeptides. Furthermore, all strains showed the production of catechol-based siderophore bacillibactin, and antibiotics bacilysocin and bacillaene. When compared for the amounts of different cyclic-peptides produced by three bacilli, B. amyloliquefaciens SB-1 showed the most noticeable amounts of all the antimicroial compounds. Plant experiment results manifested that inoculation with Bacillus spp. strains demonstrated substantial growth improvement of wheat biomass, number of spikes and dry weight of shoots and roots. The most significant results were shown by B. amyloliquefaciens SB-1 as bioinoculant over un-inoculated control plants which caused ~42% increase in the total biomass. Results of this study indicate the potential of biocontrol and biofertilizer Bacillus spp. in disease and nutrient management of crop plants, particularly cultivated in the South Asian region.

# MOLECULAR PHYLOGENY OF RACERUNNERS (*EREMIAS* FITZINGER, 1834; SQUAMATA: LACERTIDAE) FROM PAKISTAN SUGGESTS CRYPTIC DIVERSITY AND LARGE-SCALE TAXONOMIC CONFUSION

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Phylogenetic relationships among all morphologically defined species of the genus *Eremias* Fitzinger, 1834 from Pakistan were examined using mitochondrial (16S rRNA, cytochrome b) and nuclear (Rag1)markers. Overall 26 individuals of *E. acutirostris*, *E. aporosceles*, *E. cholistanica*, *E. persica* and *E. scripta* were included into the phylogeny of the genus. The classical phylogenetic analysis was conducted, resulting in concordant topologies. All species clustered into five distinct well-supported clades in the multi locus, as well as one-locus dataset. Despite variations in certain morphological characters between *E.aporosceles* and *E. acutirostris*, the former species clustered with the latter with only low inter-specific value of genetic divergence (uncorrected p-distances 1.7%). Thus, our genetic data do not support the validity of the questionable species of *E. aprorosceles*. On the other hand, *E. persica* represents a species complex with so far unknown phylogenetic clades from the Pakistani territory. To resolve the taxonomical position of this complex, geographically close species of the genus are needed to be in cluded for further analysis. *Eremias cholistanica* is phylogenetically well-supported and represents the only species of the genus distributed in the Oriental zoogeographic region. *Eremias scripta* from Pakistan turns out to be genetically distinct from the single specimen of this taxon known from Afghanistan, suggestive of another cryptic diversity in this taxon. Further research from the vast distribution ranges of *E. persica* and *E. scripta* is warranted to clarify their taxonomy and phylogeographic differentiation.

### POULTRY WASTE, A POTENTIAL BIOENERGY SOURCE: A REVIEW AND FUTURE PROSPECTS

### Huda Rehman Mir and Mushtaq A. Saleem

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Poultry industry is known to be one of the biggest organized sectors that generates huge quantity of waste. One of the challenging tasks of poultry farms is the waste disposal in order to maintain hygiene as well as improve productivity. It is most abundantly used for crop production. The excessive use of poultry litter for this purpose may cause air, water and soil pollution apart from spreading hazardous pathogenic microbes. The present review aims to assess the potential use of Poultry waste into sustainable alternative energy resource. One of the main concerns is that Poultry Industry excessively uses phosphorus as an inorganic source in both broiler and layer chicken diets that are reflected in poultry litter which causes threats of accelerating waste eutrophication. The land disposal of poultry waste and the successive environmental implications has accelerated interest into more useful and cleaner disposal options. This study also analyses the intrinsic characteristics of poultry litter as a Bioenergy fuel, as well as discusses the main thermochemical processes occurring during its conversion into energy and propose strategies to enhance its performance as a sustainable biomass. In this regard, it is imperative to find new rational alternatives for using poultry litter that results in less damage to the environment with suitable treatment to get rid of undesirable compounds and pathogens. Among the accessible alternatives, the use of poultry litter as biomass energy for heat and electricity generation through anaerobic digestion is gaining importance.

### BIODEGRADATION OF DISPERSE TEXTILE DYE BLUE-284 BY KLEBSIELLA PNEUMONIAE

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The discharge of dye-contaminated industrial wastewater into natural waterways presents a substantial risk to human and environmental health; therefore, economical and bio-friendly approaches are needed to remediate dye-contaminated wastewater from various industries before their release into the ecosystem. In this study, a local bacterial strain N4 was isolated from a textile wastewater, capable of decolorizing disperse blue 284 dye and identified as *Klebsiella pneumoniae*. The effects of pH, temperature, dye concentration, and incubation period was studied on decolorization of disperse blue 284 dye by bacterial isolate. At 20oC decolorization was 23% whereas it was 99% at 37oC and decreased to 9% at 50oC. Similarly, at pH 4, decolorization was 31% whereas 99% at 7pH. Bacterial culture showed maximum decolorization of 200ppm dye concentration, but as dye increased 400, 600, 800 and 1000ppm the bacterial potential of decolorization of dyes decreased to 79%, 78%, 51% and 34%, respectively. The decolorization percentage for disperse dyes by the strain N4 could reach 99% under optimum conditions (pH 7; temperature 37oC; 180rpm and dye concentration 200ppm for 24 hrs). Altogether, our results support the use of this bacterium in the degradation and treatment of textile industrial effluent containing azo disperse dyes.

### EVALUATION OF LIPID PEROXIDATION AND BIOCHEMICAL RESPONSE IN CHRONIC KIDNEY DISEASE PATIENTS

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Chronic Kidney Disease (CKD) is a worldwide health problem. CKD is general term for heterogeneous disorders affecting the structure and function of the kidney. Structural characteristics in CKD, loss of renal energy, and uremia result in an imbalance between free radical production and antioxidant defenses. To determine the Lipid Peroxidation and Biochemical response in CKD patients. Sixty patients of Chronic Kidney disease and fifty healthy individuals' data was taken in the study. Blood sample collected from Jinnah and Mayo Hospital Lahore. 5.0 ml blood samples were taken and subjected to centrifuge at 3000-4000 rpm for 10-15 minutes for the separation of serum. The estimation of Urea, Creatinine, GSH, SOD, AGES, NO, AOPP, and MDA were estimated. Nitric Oxide (NO) level in CKD patients was elevated remarkably (21.16  $\pm$  0.076) as compared to control (4.4  $\pm$  1.27) and statistically significant (p-value <0.00). MDA level was also increased in CKD patients (13.69  $\pm$  0.27) as compared to healthy persons (3.37  $\pm$  0.95) and statistically significant (p-value <0.00). The level of advanced oxidation protein product (AOPP) in CKD patients was decreased (0.42  $\pm$  0.26) as compared to healthy individuals (6.11  $\pm$  2.01) and also statistically significant (p-value <0.00). Present study concluded that Strong association exists between Oxidative stress, Immunosuppressant, Micronutrients and electrolyte balance in ESRD patients. Increased Lipid per oxidation leads to elevated level of MDA remarkably where as Anti-oxidants decreases.

### EFFECT OF DIFFERENT PLANT EXTRACTS ON ACETYLCHOLINESTERASE ACTIVITY OF AEDES AEGYPTI AND MUSCA DOMESTICA ADULTS

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The aim of the present study was to determine the mortality and AChE inhibitory potential of extracts isolated from three common plants i.e., Calotropis procera, Eucalyptus globulus and Mentha spicata against Aedes aegypti

and *Musca domestica*. The mortality and AChE inhibitory potentials of plant extracts were also compared with chlorpyrifos, a commonly used insecticide. WHO recommended protocol was followed for conducting bioassay tests against three different concentrations of plant extracts. The mortality rate was assessed 24 hour post treatment. The AChE activity was determined by spectrophotometry at 412 nm using Ellman's assay. Results showed that *C. procera* caused higher mortality in *Ae. aegypti* and *M. domestica* than *E. globulus* and *M. spicata*. There was a marked decline in the enzyme activities of treated groups compared to control. *M. spicata* treated groups revealed higher AChE inhibitory potential among three plants extracts. It is concluded that some plant extracts inhibit the AChE activity of insects and exhibit significant mortality in *Ae. aegypti* and *M. domestica*.

# 2. CELL AND MOLECULAR BIOLOGY, CELL BIOLOGY, GENETICS

### G9A HISTONE METHYLTRANSFERASE AS A POTENTIAL ANTICANCER TARGET FOR BREAST CANCER

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Epigenetic modifications play an important role in cancer development and metastasis. G9a is an important methyltransferase in animal cells. It controls the methylation of many cancer critical genes. Its activity is overexpressed in a wide variety of commonly known cancers especially breast cancer. Its diverse roles make it an ideal target for cancer therapy but its vital role in cell homeostasis makes it hard to target. We have used a small inhibitory molecule BIX01294 to reduce the methyl-transferase activity of G9a It is known that G9a is required for gene silencing and gene activation required for EMT which we have induced in the lab by TGFβ treatment in two breast cancer cell lines MDA-MB231 and MCF7. Through various experiments including ROS production, invasion and migration assays, cytoskeleton staining to see actin stress fiber and finally quantitative PCR for mRNA level of many EMT markers, regulator or modulators, we have concluded that G9a is more potent target for metastatic cell line like MDA-MB231 rather than for none or lesser metastatic cell lines like MCF7.

# AN IGNORED CONTRIBUTING FACTOR OF VITAMIN D DEFICIENCY DESPITE STRONG CORRELATION WITH INCIDENCE OF BREAST CARCINOMA AMONG WOMAN IN PAKISTAN

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Pakistani females are at elevated risk of breast cancer, thus there is need to explore different hereditary and nonhereditary factors. During the previous studies it has been found that vitamin-D has antitumor activity in breast cell lines with significant reduction in development of breast cancer in rats exposed to carcinogens. Women included 155 clinically diagnosed breast cancer patients from different Hospitals of Punjab, Pakistan and 247 randomly selected women as control group were recruited to find the impact of Vitamin-D deficiency in both urban and rural regions. Statistical analysis was done using SPSS version-23 to compute Pearson's Chi square test. Average age at diagnosis of breast carcinoma patients and control group at the time of interview was 44.38 ±9.8 and 45.24± 10.92 respectively. Out of total 402 subjects 190 (47.35%) of subjects were not aware of their Vit-D level. Thus, the comparison of case and control for Vit -D deficiency data, revealed P= 0.039 by performing Pearson's chi square test. Analysis in different age groups among breast cancer patients revealed that 31 to 50 years age group as most prone, with 60.9% of women presented with such deficiency. Urban women were found to have 12% greater incidence of Vit-D deficiency as compared to women living in rural areas. It is concluded that Vit-D deficiency highly contributed to incidence of breast cancer so every female must be aware about the importance of sufficient level of Vit-D and maintain it by changing their lifestyles.

### MESENCHYMAL STEM CELLS AS A THERAPEUTIC OPTION TO TREAT LIVER FIBROSIS: CURRENT SITUATION AND FUTURE PROSPECTS

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Liver fibrosis is characterized as the accumulation of extracellular matrix into liver tissues which ultimately leads to liver cirrhosis and liver carcinoma. Liver fibrosis has become the major health care issue with no obvious treatment available. Now a days, liver transplant is only treatment option which is very expensive due to organ shortage and organ rejection issues. Researchers are now moving towards cell-based therapies like use of Mesenchymal stem cell (MSCs) as a therapeutic option because of their ability to differentiate into various cell linages like cardiomyocytes, hepatocytes, osteocytes and their potential immunomodulatory properties. MSCs homes to the damaged liver and differentiate into hepatocytes thus by replacing damaged hepatic cells they promote hepatocyte regeneration and induces angiogenesis via paracrine signaling. MSCs shows great potential to repair and regenerate liver by releasing cytokinesis and growth factors. The ability to differentiate into various cell linages an immunomodulatory property of MSCs makes them a potential therapeutic option for the treatment of liver fibrosis. Still there are many challenges, like viable delivery mechanism and efficiency of homing to damaged site, which needs to be overcome in order to apply MSC-based therapy as a proper treatment.

# EVALUATION OF MALONDIALDEHYDE AND BIOCHEMICAL RESPONSE IN ORAL SQUAMOUS CELL CARCINOMA PATIENTS

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Oral squamous carcinoma cell is generally defined as an oral neoplastic disorder in the mouth. Hence, the OSCC originate from the epithelial lining, which is present in the oral cavity. Most cancers begin with lips and mouths in squamous cells, thin and flat cells that affect the lips and the oral cavity. So that's why they are called squamous cell carcinomas. The main objective of present study was to evaluate the correlation between OSCC, paclitaxel and biochemical status. 5.0 ml venous blood sample of 60 patients of OSCC treated with Paclitaxel and 50 Blood sample of Healthy individuals was taken in clotted gel vial from Oral and Maxillofacial department, Mayo hospital Lahore. Blood was further processed for the estimation of Reduce Glutathione (GSH), Catalase (CAT), Superoxide Dismutase (SOD), Malondialdehyde (MDA), Estimation of Nitric oxide (NO), Estimation of micronutrients (Vitamin A, Vitamin C and Vitamin E). The serum NO level in diseased person is  $0.46 \pm 0.62$  while in controlled healthy person is  $4.21 \pm 1.17$ . The serum NO is significant statistically (p=  $0.001 \le 0.05$ ). Level of MDA in diseased person is  $0.64 \pm 0.45$  while in controlled healthy person is  $3.18 \pm 0.11$ . Hence, MDA is significant statistically (p=  $0.000 \le$ 0.05). Level of CAT in diseased person is  $11.11 \pm 0.12$  while in controlled healthy person is  $1.31 \pm 1.08$ . So CAT is significant statistically (p=  $0.000 \le 0.05$ ). GSH level in diseased person is  $0.72 \pm 0.29$  while in controlled healthy person is  $4.00 \pm 0.41$ , so GSH is significant statistically (p=  $0.000 \le 0.05$ ). Vitamin A level in diseased person is  $10.25\pm2.16$  while in controlled healthy person is  $108.08\pm6.12$ . Vitamin A is significant statistically (p=  $0.000 \le$ 0.05). The oral squamous cell carcinoma is very pronounced topic. In this cohort study, we found a strong relationship and correlation of lipid peroxidation and biochemical response in patients, receiving paclitaxel. OSCC is very common among all the cancers in the world.

### STUDIES ON INDUCTION OF NUCLEAR ABNORMALITIES IN PERIPHERAL BLOOD ERYTHROCYTES OF *LABEO ROHITA* EXPOSED TO MERCURY

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Heavy metals are expanding due to anthropogenic activity in aquatic environment. These metals ingress into food chain and cause genetic mutilation to aquatic fauna. During current study, acute toxic effect of mercury was determined in *Labeo rohita* under manipulate PH, total hardness and temperature of water by using static bioassay method. Micronucleus test was used to recognize the genotoxic effect of mercury on fish in the lab. The acute toxicity in terms of mercury 97-hour LC50 and lethal concentration (97-hr exposure period) of mercury was calculated ass 146.92±0.02, 294.97±0.01mg/L, respectively for *labeo rohita* by using prohibit analysis method. After acute trial, fish was exposed to two sub lethal concentration of mercury (HgT1 and HgT2) separately. After chronic exposure of mercury, the blood samples were taken from the caudal vein of each species and processed for analysis of structural nuclear anomalies (%) i-e lobbed, blabbed, dumbbell, deshape and binuclei in the peripheral blood cells of *labeo rohita* in dose dependent manner. The group exposure to metal display significant variation in cellular abnormalities and frequency of DNA damage. Conclusion revealed concentration dependent increase in the frequency of micronuclei and other nuclear aberration.

### IDENTIFICATION OF GENETIC DIVERSITY AMONG THE SNAIL SPECIES USING MOLECULAR MARKERS

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Snails are mollusks fitting to the class Gastropoda. The members of gastropoda, slugs and snails make up 80% of all mollusks. The gastropods live all over the world, from the Arctic and Antarctic waters to the equatorial areas. Snails are the most successful and second large group of invertebrates. Marine mollusks are best to study genetically. They play a role of intermediate host in diseases affecting human and livestock. It's important to study their genetic diversity to reduce the disease ratio. The molecular markers are important to understand the gastropods diversity along with their morphological characters. RAPD analysis will be used for genomic characterization of snails. RAPD markers are important in different gene studies. RAPD analysis will be accomplished by PCR based amplification. The purpose of this study is to check genetic diversity among snail species and their molecular characterization by using RAPD markers. Analysis revealed that Close association was observed among the species of genus Physa and these species were found to be closely clustered in one group while the species *Physa fontinalis* and *physa gyrina* is distantly related with *Physa acuta* in this group showing it to be genetically distant from the same group. The cluster analysis shows *that Physa acuta* lies separately in cladogram, as it might be a new species. The maximum similarity matrix is 94% and minimum genetic similarity is 61%. Polymorphism revealed by the RAPD fragments is 71.4%.

# MOLECULAR EVALUATION OF HEREDITARY AUTOSOMAL RECESSIVE WOOLLY HAIR/HYPOTRICHOSIS FROM DISTRICT KARAK, KHYBER PAKHTUNKHWA, PAKISTAN

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Hair diseases are primarily associated with the follicles of hairs. These may refer to excessive shedding or baldness. Various factors contribute to these disorders e.g. genodermatoses is responsible for hypotrichosis, alopecia

areata occurs due to autoimmune disorders and androgenic alopecia is caused by hormonal imbalances. Hereditary hair disorders in humans are classified into syndromic and non-syndromic forms. In syndromic forms, hair symptoms appear as part of a genetic syndrome that can exhibit other skin symptoms and/or various systemic manifestations, whereas in non-syndromic forms, affected individuals only show hair phenotype. Autosomal recessive woolly hair/hypotrichosis (ARWH/H) is a non-syndromic hair irregularity described through short and firmly curled scalp hairs at birth which can lead to sparse hair later in life. In this hair growth abnormalities manifest as either hypertrichosis (excess of hair) or hypotrichosis (lack of hair). Until now seven autosomal dominant and equal numbers of autosomal recessive forms of isolated hair loss disorders have been mapped on different human chromosomes. In the present study we report, 3 unrelated consanguineous Pakistani families having multiple affected individuals. Linkage in these families was searched by genotyping microsatellite markers. Two of the families showed linkage to the LAH3 locus on chromosome 13q14.11-q21.32. These families were then subjected to direct sequencing of the LPAR6 gene, which encodes a G protein-coupled receptor. Sequence analysis of the LPAR6 gene showed a recurrent missense mutation (p. G146R) in affected members of the families. Sequence analysis disclosed same pathogenic sequence variation at nucleotide position (c.436G>A), replacing Glycine amino acid with Arginine (p. Gly146Arg) in both families (A, B). The identification of a genetic defect in LPAR6 suggests that this enzyme regulates hair growth. Our study increases the spectrum of known LPAR6 mutations and highlights the importance of this receptor in human hair growth and texture.

### GENETIC ANALYSIS OF HEREDITARY ALBINISM PATIENTS FROM DISTRICT TANK, | KHYBER PAKHTUNKHWA, PAKISTAN

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Albinism is a combination of several types of inherited disorders in which the melanin production is reduced or totally absent. Clinically, albinism is categorized into oculocutaneous albinism (OCA) and ocular albinism (OA). Until now seven genes namely TYRP1, C10ORF11, SLC24A2, SLC45A2, TYR, MCIR and OCA2, have been identified for causing non-syndromic OCA. Whereas, several genes namely MYO5A, HPS3, HPS4, DTNBP1, BLOC1S3, PLDN, LYST, HPS5, HPS6, RAB27A, MLPH, LYST, HPS1 and AP3B1 have been identified for causing syndromic OCA. In Pakistan, families having OCA disorders are significant hotspots for hereditary studies and therefore diagnostics and hereditary procedures is needed. The present study aimed at identification of novel genes/loci or sequence variants in the OCA families in Pakistan and their effects on the individuals. Three Pakistani OCA families were recruited from district Tank, Khyber Pakhtunkhwa, Pakistan. In the current study all the three OCA families processed for mutational identification, showing linkage to OCA2 gene. Two families (A and B) showed deletion mutation in exon 19. In family (C) ten exons (4, 10, 13, 14, 15, 16, 19, 21, 23, 24) were PCR amplified, as mutations in these exons are reported in the Pakistani (Pashtun) population but no sequence variant was found. It suggests that mutations in other exons may be responsible for OCA in the studied family, which will require further studies.

### MOLECULAR ANALYSIS OF HUMAN HEREDITARY SPLIT HAND AND FOOT MALFORMATION PATIENTS FROM DISTRICT KARAK

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Split hand and foot malformation (SHFM) is a congenital, multifaceted inherited rare limb developmental disorder including deep median clefts of the hands and feet, and aplasia and/or hypoplasia of the phalanges. The

causes of split hand and foot malformation were known to be associated with mutations in multiple genes (TP63, DLX5, ZAK, FGF8, FGFR1, DLX6, EPS15L1, BHLHA9 and WNT10B) involved in limbs development and different regulatory pathways. To date, it has been reported that 9 loci including SHFM 1-6, SHFMLD3, chromosome 8q21.11-q22.3 and chromosome 19p13.11 are involved in SHFM. However, only five genes (DLX5, DLX6, WNT10B, TP63, EPS15L1) associated with SHFM have been recognized so far. The current study aims to identify novel genes / loci or sequence variants in Pakistani SHFM population. Three Pakistani SHFM families (A, B, C) were sampled from district Karak and were processed for identification of mutation. Family A showed linkage to EPS15L1 gene, where a Single nucleotide polymorphism (SNP) was found in WNT10B gene. No linkage was found in family B. Family (C) followed autosomal dominant mode of inheritance, exons (3, 4, 5, 7, 8,13) in TP63 gene, exons (1, 2, 3) in DLX5 gene and exons (2, 3) in DLX6 gene were PCR amplified but no sequence variant was found. Since no mutation was found in the exons of the above-mentioned genes, it indicates that mutations in other exons or other genes in the studied family may be responsible for SHFM, which will require further studies.

# POTENTIAL ROLE OF GENOMICS FOR TREATMENT OF HEARING LOSS AND DEAFNESS

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Deafness is partial or total inability to hear influencing one individual in every 500 babies and 278 million adults around the world. It can be caused due to genetic and environmental factors, while the exact etiology is unknown. Genetic deafness is 70% non-syndromic, inherited in mostly an autosomal recessive pattern, autosomal dominant or X-linked pattern of inheritance. Deafness can also be the result of mitochondrial impotence. Around 34% genes out of 102 loci are supposed to cause genetic deafness. Current advancements in genomics have introduced new patterns of understanding the inheritance of deafness. Induction of induced pluripotent cells, cell replacement, cochlear bone improvement and gene therapies are the clouds with silver linings and are considered a huge breakthrough in genomics for the cure of deafness. One of the main focus of the otology researchers is to deliver a gene in to the inner organ of the ear to restrain the hearing loss. Although these therapies are in experimental stages and are expected to be perilous, but if successful, geneticists may be able to cure genetic disorders.

# DETECTION AND PHYLOGENETIC ANALYSIS OF CYTOCHROME B GENE OF LEISHMANA TROPICA IN PATIENTS OF DISTRICT KARAK, PAKISTAN

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Leishmaniasis is a group of wide spread zoonotic and anthroponotic disease caused by a variety of different species of the genus *Leishmania* that infects humans and other vertebrates. In Pakistan, *Leishmania tropica* is considered as a causative agent of cutaneous leishmaniasis (CL). The current study was conducted from May 2019 to January 2020 in highly epidemic area of the KP. A total of 60 (100%) microscopic confirmed positive samples were collected from Karak health centers. Initially molecular detection was carried out for microscopically confirmed CL by simple Polymerase Chain Reaction (PCR) and then the amplified PCR product was used to know the sequence analysis of *Cytochrome b* gene (*Cyt b*) to investigate the presence of genetic polymorphisms among *L. tropica* isolates. PCR assay targeted *Cyt b* using specific primers and detect *leishmania tropica* in which 52 (86.66%) was confirmed by PCR products and 40 (66.66%) was completely confirmed by sequence analysis to investigate genetic

polymorphism. However in the present study, the sequence analysis of the amplified *Cyt b* will be applied to investigate the presence of genetic polymorphisms among *L. tropica* isolates and correlate the findings with the clinical features of CL lesions in patients in southern region of Khyber Pakhtunkhwa. Therefore, the current study has been planned to further elaborate the phylogenetic relations of *leishmania tropica* in the endemic area of Pakistan.

### HEMATOLOGICAL AND BIOCHEMICAL STATUS OF BETA-THALASSEMIA IN PAKISTANI AND AFGHANI PATIENTS OF QUETTA CITY, PAKISTAN

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Thalassemia is a genetic blood disorder in which body is unable to synthesis hemoglobin characterized by chronic anemia. Improper erythropoiesis is the major problem in thalassemia. 50 Pakistani and 50 Afghani patients (male & female) from various public sector hospitals of Quetta city for the period of one year, were include in this study. Patients were divided into four groups (N=25 each group): Group-I: Included male and female Pakistani control individuals, Group-III included male and female Afghan control individuals, Group-IV included male and female Afghan thalassemia patients. BMI (Kg/m²) was recorded. 3-5 ml of blood was collected, serum was isolated and biochemical analysis for hematological parameters (Hb, MCV, MCH, MCHC, PCV), renal function test (urea, creatinine) and liver function test (AST, ALT) were done. A significant reduction in the BMI (P<0.0001) was reported both in Pakistani and Afghani thalassemia patients as compared to normal individuals. Significant decrease was found for hematological parameter in thalassemia patients in both population and in both genders. Serum AST, ALT, creatinine and urea was significant increase in both Pakistani and Afghani thalassemia patients as compare to Control. The prevalence of thalassemia is more severe in Afghani patients as compare to Pakistani patients since in Afghanistan health facilities are very poor, inter tribe marriages are very common and lack of knowledge.

# CYTOCHROME B BASED GENETIC BARCODING OF CHIROPTERAN SPECIES FROM FATA, KPK, PAKISTAN

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Bats (order chiroptera) constitutes almost a quarter of all known mammalian species of the world. Species identification is a pre-requisite to take conservation measures. Moreover, bats carry significant ecological and environmental importance. Asian chiropteran fauna, with particular reference to Pakistan is poorly studied. Herein for the current investigation, sequence data of bats from FATA region of Pakistan is provided as a foremost record. This study explored the genetic diversity in bats from FATA using a robust molecular characterization technique. The technique uses sequencing of cytochrome b gene (cyt b), present on the mitochondrial genome, for subsequent genetic barcoding. Genetic analysis results revealed that FATA bats belonged to the following genera - *Scotophilus*, *Rousettus*, *Pteropus*, *Pipistrllus* and *Eptesicus*. Furthermore, phylogenetic comparison of FATA bats with other Asiatic bats was conducted to explore closely related species. The data will enable researcher to build an improved evolutionary landscape of chiropteran from this region.

### IDENTIFICATION OF NOVEL SNPS OF CAST GENE IN THREE DIFFERENT CATTLE BREEDS OF PAKISTAN

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Calpastatin is the variable component of calpain system, encoded by CAST gene. In skeletal muscle growth and post mortem tenderness of meat there is an essential role of calpastatin. Cast gene is located on bovine chromosome 7. Its main purpose is to change the proteolytic action of enzymes reliable for post-mortem myofibril deterioration. Present study was aimed at identification of novel SNPs of Calpastatin gene in three different cattle breeds of Pakistan. In this study PCR-Gel Electrophoresis and DNA sequencing techniques were used for obtaining the results. In total 7 mutations were identified with the help of PCR-Gel Electrophoresis and DNA Sequencing technique. Based on Genetic code, we have identified (02) mutations in Exon-5 of Red Sindhi cattle breed including a silent mutation at 80 bp in which CAA(Glutamine) codon was changed to CAG(Glutamine) but does not causes any change of Amino acid hence will not effect on meat qualities of the Red Sindhi cattle, Another mutation was also found in Exon 5 of the Red Sindhi cattle breed, which causes change of codon from TGT to CGT, in which former was coding for the Cystine and later is coding for Arginine Amino acid change from non-essential to essential. In Sahiwali cattle, 02 mutations were also identified in exon 3, in which AAA codon was changed to ACA at 27 bp, which results in substitution of Lysine to Threonine amino acid. Second mutation caused the change of codon from ATC to AGC which results in change of amino acid from Isoleucine to serine amino acid. We have also identified 03 mutations in Exon5 of Tharparker cattle breed. First one was identified at 35 bp of Exon 5, in which GTA codon was changed to GTG, which does not cause any change of amino acid so called silent mutation (Valine to Valine). Second mutation was identified at 57 bp in which GAA codon was changed into AAA, which results in change of Glutamine amino acid to Lysine Amino acid. Third mutation resulted in change of TAT codon to TGT codon that caused change of Thyronine amino acid to Cysteine amino acid. Based on this study we suggest that Exon 5. Essential mutation can be effect the meat properties in position manner and hence can be used in Marker Assisted selection of this cattle breed.

# IDENTIFICATION OF GENETIC DIVERSITY AMONG THE BUFFALO SPECIES USING MOLECULAR MARKERS

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Pakistani buffaloes with chromosome number of 50 belong to river type. These animals have great tolerance against variation in quality and quantity of feed resources and have major contribution in the economy of Pakistan providing draught milk and meat. Almost 37.7 million buffalos are present in our country, out of which 49% are present in Punjab. Classification of native buffalo breeds on the base of genetic variation is important for making breeding and genetic conservation policies. For genetic characterization different molecular markers are used but microsatellite markers are more reliable for genetic study. The purpose of this study is to check the genomic variation among the buffalo species and their characterization at molecular level by using RAPD markers. The result showed close association among *Ravi* and *Nili-Ravi* while the *Nili* specie showing it to be genetically distant from them. The average observed heterozygosis values were 0.58 and 0.69. The maximum similarity matrix is 0.92% and minimum genetic similarity is 0.87 %. The three species of buffalo population showed less genetic diversity as indicated by an Fst value (0.05), 74.87% polymorphism revealed by RAPD fragments.

# APPLICATION OF BIOINFORMATICS TOOLS FOR THE MOLECULAR TAXONOMY AND THE APPRAISAL OF GENETIC DIVERSITY IN COMMERCIALLY IMPORTANT PORTUNID CRABS FROM THE COASTAL WATERS OF PAKISTAN

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The expansion of DNA technology directed to the generation of huge amounts of data from the plant and animal's genome. The main source of marker is mitochondrial DNA (Mt-DNA), mini satellite, microsatellite and nucleotide sequences assay using highly specific PCR primers. Bioinformatics is recognized as part of the crucial knowledge base of frequently career paths in biomedical research and healthcare. Nonetheless, there is little agreement in the field over what that knowledge entails or how best to provide it. Application of Bioinformatics tools, to explore genetic diversity through new molecular markers is a constant challenge for the progress in phylogenetic research and in particular, to study the genetic variation in organisms. A major contribution comes from molecular marker used for molecular taxonomy and analyzing genetic variation. During the current research the online available Bioinformatics tools, i.e. BLAST for DNA or protein sequence based on similar identity, ORF to find Open Reading Frame (ORF) and diverse molecular genetics software are being used to characterize the mitochondrial DNA analysis of noncoding16S rRNA and coding gene COI for the nucleotide and haplotype diversity, inter and intra specific genetic divergence, and phylogenetic relationship of Portunid crabs from the coastal waters of Pakistan. In the present study 14 species of family Portunidae were identified and their phylogenetic relationship was also strongly support polyphyletic nature of Portunus as studied through recent molecular data in comparison with previous conclusions.

#### GENETIC DIVERSITY OF SOME FLATFISHES FOUND ALONG THE COAST OF PAKISTAN

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Sole fishes belong to order Pleuronectiformes and generally well recognized for their specific body structure as compressed asymmetrically with flattened body shape additionally, eyes are present on one side of the fish's body. The eye side is usually remarkable pigmented whereas the blind side of the body is de-pigmented. Sole fishes are also the conspicuous element of estuarine habitat on Pakistan coast and some species are evident in the fisheries catch with significant contribution. Along the coast of Pakistan, the Cynoglossidae family recognized as a diverse in a number of species as nine representative species, whereas Soleidae family contains six species as a contrary, Paralichthyidae and Bothidae contains five species of sole fishes. Nonetheless Psettodidae and Samaridae family contains one species of sole fish (FAO, 2015). The current study is based on genetic diversity of sole fishes which were collected from five different fish harbors (i.e., Karachi, Korangi, Keti Bandar, Ormara, and Pasni) along the coast of Pakistan. The genetic diversity was studied through 16S mtDNA gene sequence. A total fourteen specimens of seven species were selected for the study. The selected gene was amplified through PCR and ranges from highest base pair in *C. arel* fish that belong Cynoglsidae family and lowest base pair was measured 525 from the species of Pralichthyidae family. The procured sequences were submitted to Gene Bank for the record.

#### GENETIC STATUS OF HYPOPHTHALMICHTHYS MOLITRIX IN INDUS RIVER AS ASSESSED BY MOLECULAR MARKERS

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The genetic integrity of fish populations has been continuously threatened due to various anthropogenic actions. These losses in genetic diversity present an erosion of global biodiversity. The purpose of current study was to assess the genetic status of Hypophthalmichthys molitrix in Indus River as assessed by molecular markers. For the purpose, sampling was done from five different sites viz. Chashma Barrage, Taunsa Barrage, Jinnah Barrage, Ghazi Ghat and Guddu Barrage. Samples were stored in crushed ice and transported to Aquaculture Biotechnology Lab, University of Agriculture, Faisalabad. For DNA isolation, muscle tissues were taken from dorsal portion of fish and isolated by following the standard "proteinase-K and phenol/chloroform" method. 0.8% agarose was used to check the quality of isolated DNA while quantity was assessed by using nanodrop. Genomic DNA was PCR amplified by Labeo rohita cross-species amplification of H. molitrix by using five primers viz. Lr-12, Lr-21, Lr-22, Lr-38 and Lr-40. Amplified PCR products were resolved on a 5% non-denaturing PAG containing 19:1 acrylamide: bis-acrylamide and visualized by silver-staining method. Data were analyzed by using different softwares including TFPGA, FSTAT and POPGENE. The results showed low-to-moderate level of genetic diversity. The number of alleles on each locus ranging from 2.0 to 6.0 with an average 3.48 was observed at various loci. Average observed and expected heterozygosities ranged from 0.664 to 0.76 and 0.631 to 0.664, respectively. For all tested loci, population combinations showed significant deviation (p<0.05) from HWE. The AMOVA indicated most of the variation lied within individuals in populations of H. molitrix. Based on Nei's genetic distance, UPGMA dendrogram was constructed that resulted in two clusters: one cluster consisted of JB and CB while other consisted of GG, GB and TB. The findings of this research will be useful for restoring, conservation and monitoring the natural and farmed aquatic fish species in Pakistan.

# DISTRIBUTION AND EVALUATION OF POLYMORPHISMS IN HUMAN IL-18 GENE PROMOTER ASSOCIATED WITH VIRAL CLEARANCE IN HCV INFECTED PATIENTS IN KHYBER PAKHTUNKHWA, PAKISTAN

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Interleukin-18 is a pro-inflammatory cytokine which plays important role in anti-viral immune responses by augmenting the action of certain immune cells. Two promoter polymorphisms in gene sequence of IL-18 at site -607C/A and -137G/C have been found to be associated with delayed hepatitis C virus (HCV) clearance and persistence. The present study is aimed to examine the frequency of these promoter polymorphism and to determine their role in HCV outcomes in HCV infected individuals in Peshawar. A total of 70 HCV infected were considered for the study and divided into two groups; Group 1 consisting of Anti-HCV positive and HCV RNA negative patients(n=40),and Group 2 including patients positive for both Anti HCV and HCV RNA (n=30), 30 healthy individuals with no HCV infection were also included in the study. Blood (5ml) was taken and analyzed first through PCR for detection of HCV RNA, and the genotypes of IL-18 promoter region was amplified through PCR, which were further analyzed through restriction fragment length polymorphism (RFLP) for detection of polymorphism. The overall frequency distribution of these single nucleotide polymorphisms at position -607 comprised 62% (62/100) of allele A and 32% (32/100) of allele C while the most prevalent allele at loci -137 was allele G (59%). The -607 A allele was more common in clearance group (Group 1) than in persistence group (Group 2). The haplotype AC (-607A &-137C) was found to be most frequently linked with HCV clearance in study population. No significant difference was observed in gender wise distribution of these polymorphisms. This study indicates that these variations in promoter of IL-18 gene might have a significant role in pathogenesis and outcome of HCV infection in different populations in Peshawar.

### GENETIC CHARACTERIZATION OF ROHU (*LABEO ROHITA*) POPULATIONS IN RIVER JHELUM BY USING DNA MARKERS

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Genetic variation in a species enhances the capability of organism to adapt to changing environment and is necessary for survival of the species. Anthropogenic changes to the environment are one of the major causes of species decline worldwide. The present research was conducted to assess the genetic status of Labeo rohita form five different sites of River Jhelum (Head Trimmu, Mangla Dam, Rasul Barrage, Jhelum Bridge and Pind Dadan Khan) using microsatellite DNA markers. Total 30 samples were collected from each site then preserved for transport. For the extraction of DNA Phenol, chloroform and isoamyl alcohol method was used and confirmed by 0.8% agarose gel. Five polymorphic microsatellite loci (Lr12, Lr21, Lr22, Lr38 and Lr40) were utilized for analysis of genetic variation. The genetic diversity in terms of heterozygosity was observed moderate in all examined populations of L. rohita. The highest mean value of observed heterozygosity was found 0.833 in MD population and the lowest 0.520 in JB population. The estimated heterozygosity was ranged from 0.636 to 0.672 for JB and RB. Population of MD showed the highest  $F_{is}$  value -0.307 and lowest 0.182 observed in JB. The highest level of genetic differentiation ( $F_{si}$ ) was found 0.196 in JB-PDK while, the limited 0.0012 between the populations of HT and PDK. The AMOVA showed that 7.154% variation was contributed due to the variation between populations of L. rohita in this study. The largest value of Gene flow (Nm) was observed 20.9777 at locus Lr38 while the lowest value of Nm was noted 7.1717 at locus Lr21. Genetic relatedness populations was investigated by constructing UPGMA dendrogram which showed the same clusters had a close genetic relationship. STRUCTURE HARVESTER admixture model inferences showed highest estimated log-likelihood mean value and delta-k value for K =3. The information about L. rohita genetics form this study will be helpful for effective management of wild populations.

#### GENETIC CHARACTERIZAITON OF CYTOCHROME C OXIDASE SUBUNIT 1 GENE OF ECHINOCOCCUS GRANULOSUS IN LIVESTOCK OF DISTRICT BANNU

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Cystic Echinococcosis is a serious helminthic zoonotic infection caused by the tapeworm, Echinococcus granulosus. The disease is highly prevalent in the areas where humans, dogs and cattle reside in close proximity. On the basis of genetic examination there are 10 genotypes of Echinococcus granulosus. The current study was designed to evaluate the presence of the parasite in livestock and identify the active genotypes of E. granulosus in district Bannu of Khyber Pakhtunkhwa. A total of 1246 animals were examined and 112 hydatid cyst samples were collected. The overall prevalence of CE was found to be 8.99% among the examined animals. CE was highly prevalent in cows 59.82 %, followed by sheep 20.53%, buffaloes 13.39, goats 6.25%. Gender-wise distribution revealed that females were more likely to be infected with CE 14.7 % than males 4.7 %. Infection rate was increasing from low to high with the increasing age of animals. Among the 112 hydatid cyst, 54 (48.21%) were found in livers, 48 (42.85%) in lungs, and only 10 (8.92%) cysts were found both in liver and lungs of the infected animals. For molecular analysis, genomic DNA was extracted from the fluid of fertile cysts and subjected to PCR targeting mitochondrial cytochrome c oxidase subunit 1 (COX-1) gene. The PCR amplified products were sequenced and phylogenetic analysis was performed. Phylogenetic analysis revealed E. granulosus genotype 1 (G-1) in all of samples. The study confirms the presence of E. granulosus (G-1) in the study area and that the livestock may have a prominent role in transmission dynamics of the parasite.

### GENETIC POLYMORPHISM OF ECHINOCOCCUS GRANULOSUS BASED ON MITOCHONDRIAL NADH DEHYDROGENASE SUBUNIT 1 GENE

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Cystic Echinococcosis (CE) is a serious zoonotic and pathologically important helminthic infection caused by hydatid cyst of *Echinococcus granulosus*. It is highly endemic in the herd keeping areas of the world including Pakistan. The current study was designed to assess the presence of the parasite in livestock and recognize the active genotype (s) of *E. granulosus* in Khyber Pakhtunkhwa, Pakistan. A total of 1,587 animals, including cows, buffaloes, goats, and sheep, were examined and 135 hydatid cyst samples were collected. The overall prevalence of CE was found to be 8.5% among the examined animals. CE was highly prevalent in cows 61.5%, followed by sheep 22.2%, goats 11.1%, and buffaloes 5.2%. Gender-wise distribution revealed that females were more likely to be infected with CE 11.11% than males 6.41%. CE infection was found to be highly prevalent 17.4% in older age groups of animals than youngers 1.1%. Among the cysts, 55.5% were found in livers, 37.0% in lungs, and only 7.4% cysts were found both in liver and lungs of the infected animals. For molecular analysis, partial sequence of mitochondrial NADH dehydrogenase subunit 1 (NADH-1) gene, was used. The obtained sequences were aligned and compared with the reference sequences retrieved from National Center for Biotechnology Information (NCBI). Phylogenetic analysis revealed *E. granulosus* genotype 1 (G-1) as the prevailing genotype in the study area. The study confirms the presence of *E. granulosus* (G-1) in the study area and that livestock may have a prominent role in transmission dynamics of the parasite.

#### GENETIC DIVERSITY OF *OREOCHROMIS NILOTICUS* FROM RIVER INDUS AS REVEALED BY SSR MARKERS

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Genetic deterioration of aquatic resources due to anthropogenic activities has become a challenge for conservation biology in last few decades. The proposed study was performed to evaluate the molecular diversity of Oreochromis niloticus from different sites of River Indus. A total of thirty individuals of appraisal fish were collected from each selected site (Ghazi Ghat, Chashma barrage, Jinnah barrage, Taunsa barrage and Guddu barrage). Genomic DNA isolation was carried out from dorsal muscles of fish. The qualitative and quantitative assessment of the DNA was carried out by electrophoresis and spectrophotometry and target microsatellite loci were subjected to PCR amplification using 5 species-specific polymorphic primers. Amplicons were separated by using PAGE and the screened allelic bands were scored manually. For the measurement of indices of fish population genetics, various analytical tools such as FSTAT, TFPGA, POPGENE, ARLEQUIN and STRUCTURE were used. Highest level of genetic diversity was observed in Chashma Barrage population. Individuals of Guddu Barrage were found to be genetically less related to Ghazi Ghat population while more closely related to Tarimu population. Values of allelic richness over different loci were ranging from 9.846 to 10.366. The values of  $F_{IS}$  ranged from 0.122 to 0.244. The average Ho values ranged from 0.6533 to 0.7667 for all examined populations. Genetic differentiation was highest between Guddu Barrage and Jinnah Barrage populations. It was concluded that the genetic diversity of O. niloticus was low to moderate at River Indus due to little gene pool as compared to its native populations. This study will be helpful for making new strategies regarding the effective management of O. niloticus for conservation of genetic resources as well as biodiversity.

#### SCREENING OF HUMAN BLOOD FOR THALASSEMIA DISEASE IN SWAT VALLEY

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The aim of our study was the screening of human blood for thalassemia disease in the students, University of Swat. For this purpose blood test were performed to screen out different types of cells such as teardrop, elliptocyte, sickle cell, macrocyte and anemic blood. During this study a total of 318 blood samples were collected from asymptomatic population of university of swat, whose belongs to different localities of Khyber Pakhtunkhwa during this study, 118 samples was unclear, and the remaining 200 was clear for the screening through microscopy, out of which, 26 samples were normal while the remaining 174 samples found thalassemic cells i-e macrocyte, tear drop, Elliptocyte, Sickle cell, Macrocyte, Anemic blood. The percentages of this cell are: macrocyte 1.1%, teardrop 30.1%, elliptocyte 23.4%, sickle cell 3.9%, anemic blood 41.2%. These all cells are the signs of thalassemia Splenic abnormalities, megaloblastic anemia, iron deficiency anemia, hemolytic anemia, jaundice, splenomegaly, vitamin b-12 deficiency, folate deficiency, liver disease, hypothyroidism, blood lose and faulty red blood cell production.

# GENETIC ANALYSIS OF UBIQUITIN C REDUCTASE COMPLEX CHAPERONE (UQCC) GENE IN THREE CATTLE BREEDS PAKISTAN

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The identification of mutations in candidate genes involved in genetic traits of specific interest remains a challenge, significant progress has been made in this area in recent years. Several strategies to break the bottleneck barrier of information have been developed or are being developed. In this study, we have identified 05 mutations in the exon 1 region of the UQCC gene in the three cattle breeds, namely the Dhanni cattle breed (02 mutations), the Red Sindhi cattle breed (02 mutations) and the breeds Sahiwali cattle identified a total of 01 mutations with the help of PCR-Gel electrophoresis and DNA sequencing technology. According to the genetic code, the identified mutations were classified in Silent Mutation in the Dhanni cattle breed (01) and in Elimination Mutation (01). Similarly, 01 mutation of elimination of the breed of Sahiwali cattle was eliminated. However, we have identified 01 nonsense mutation in Red. The Sindhi cattle breed, which causes the replacement of non-essential amino acids in essential amino acids, could be useful for breed mixing and breeding technology for marker-assisted selection.

#### REVERSE GENETICS OF DROSOPHILA MELANOGASTER CYCLOPIA MODEL

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Cyclopia is a rare and fatal congenital anatomical anomaly resulting in the disproportional cleavage of right and left hemisphere of the brain, characterized by the presence of single eye and/or partially divided eye in the new born. We have developed a model of cyclopia by continuous incestuous breeding in *D.melanogaster* at DUHS. The aim of this investigation is to explore the underlying genetic basis of cyclopic fruit fly via reverse genetics. Briefly, genome mining has shown existence of two sequence specific orthologues of human *SHH* and *PAX6* genes in the *D.melanogaster*. To further the investigation full length molecular models of human SHH and PAX6 and respective

orthologues in D.melanogaster hh and pax6 were developed by a composite approach entailing iterative threading, homology modelling and ab initio modelling. The models were refined and assessed for both thermodynamic and structural attributes. The sequence and structural comparisons showed noticeable degree of conservation between both human and D.melanogaster orthologues pointing to the functional similarities between respective orthologues. Consistently, Ca backbone superimpositions of respective homologues also represent strong back bone conservation. However, subtle but important variations were observed in the spatial positioning of the side chains of certain residues. This may in turn lead to the functional divergence between these molecules. In summary the data represent that the anatomical similarities of cyclopia between humans and D.melanogaster has conspicuous molecular and/or genetic basis. The findings than could be then exploited to strategize preventive and/or correctional measures by gene editing methods such as CRISPR.

# A RECURRENT MISSENSE MUTATION IN THE *EDAR* GENE CAUSES SEVERE AUTOSOMAL RECESSIVE HYPOHIDROTIC ECTODERMAL DYSPLASIA IN TWO CONSANGUINEOUS KASHMIRI FAMILIES

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Hypohidrotic ectodermal dysplasia (HED) is a rare congenital disorder arising from the abnormal development of ectoderm derived structures including skin, hair, nails, teeth and glands. These patients have sparse hair on the whole body including scalp and hypoplastic teeth. They have no resistance to heat due to abnormal sweat glands. A total number of four genes namely ectodysplasin A (EDA), ectodysplasin A receptor (EDAR), EDAR-associated death domain protein (EDARADD) and Wnt family member 10A (WNT10A) are known to be involved in the etiology of HED. In the present study we have ascertained two large multigenerational Kashmiri families (A and B) with autosomal recessive form of HED. By using whole exome sequencing and different bioinformatics tools we detected We identified a rare (MAF 0.00007) homozygous missense a recurrent mutation causing severe HED. (chr2:109513410:(hg19);NM 022336 c.1300T>C;p.W434R) mutation in exon 12 of EDAR (2q11-q13) gene. This variant segregated with homozygous form in all patients and their obligate carriers were heterozygous in both families. Exclusion of more than 100 ethnically matched controls in-silico analysis predicted this variant as "damaging" and thus were assumed disease causing. Identification of the same homozygous mutation segregating with disease in two different families supports the gene's important role in the development of the disorder and it may contribute to novel approaches for the prenatal diagnosis and genetic counseling of families with EDAR related disorders.

### 3. HUMAN AND ANIMAL DISEASES

# ETIOLOGY AND RISK FACTORS ASSOCIATED WITH INTELLECTUAL DISABILITY IN PATIENTS OF PUNJAB PAKISTAN

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Intellectual disability (ID) is characterized as mental retardation. It is characterized by person's abilities impairment at the time of embryonic development. The main objective of this study is to explore ID causes and its prevalence in the Lahore population. To avoid severe or profound ID early diagnosis, clinical evaluation, genetic counseling, and proper treatment is necessary. A proper guidance must require for the parents to admit their ID children in respective special school. A retrospective, analytical study was performed from September 2018 to June 2019 on Intellectual disability patients by visiting different hospitals of Lahore. Patients from different cities of Punjab including Lahore, Faisalabad, Narrowal, Wazirabad, Gujranwala, Kasur, Gujrat, Sialkot and Manawa visited these hospitals. In this study etiology and risk factors of intellectual disability (Age, Gender, IQ level. Environmental influence, Genetic disorder, Perinatal infection, Neonatal infection, Postnatal infection, physical abnormalities) were studied. The overall prevalence of ID was 7.5% (120/900) and among which 62% were males and 58% were females. Cases of ID were observed in different age groups but the most prevalent age group was 1-10 years 48.33%. The prevalence of Mild, moderate, sever and profound ID was about 53.33% (n=64), 30.83% (n=37), 14.17(n=17) and 1.67% (n=2) respectively. Social deprivation and malnutrition were the major causes of ID. Genetic and other factors were also related to ID. This present study illustrates an association of different Genetic as well as environmental factors with Intellectual disability and it also enhances our understanding about the prevalence of different parameters of ID.

### DIABETIC RETINOPATHY (DR) IN DIABETES MELLITUS TYPE 1(T1DM) PATIENTS OF DIFFERENT AGE GROUPS IN HYDERABAD AND JAMSHORO

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Diabetes mellitus (DM) is big threat towards public health. In 2013 >382 million people suffered from DM. The T1DM is autoimmune disorder and T1DM leads due to many factors i.e., environment, heredity etc. Patients with DM are leads to be diabetic retinopathy (DR). The number of DR patients' increases due to increasing the prevalence of DM. DR is a common complication of DM patients; DR is caused by chronic damage to the small blood vessels in the retina. Most common complications of T1DM are DR, representing the leading cause of blindness in young adults. The aim of this study was to evaluate the frequency of DR of different age group patients with diabetes mellitus type 1(T1DM) and was conducted in medical outdoor patient (OPD) of two main hospitals named as Civil Hospital Hyderabad and LUMHS Hospital Jamshoro from July 2018—December 2018. The results found that out 180 DR patients were enrolled in the present study. Out of 180 patient DR 120 (66.66%) males and 60 (33.33%) females. Out of 180 DR patients, 117 (65%) patients have NPDR & 63(35%) have PDR. Out of 117 NPDR, 50 patients were mild NPDR, 35 patients were moderate NPDR and 32 patients were severe NPDR. Of the total DR patients, majority 100(55%) had 15-20 years of duration of T1DM, followed by 60 (%) 10-15 years and 20 (%) up to 10 years. The results found that the maximum number of patients of DR were at the age 26–30 years 99 and 81 patients of 20-25 years of age. Out of 180 DR patients 60 have positive family history and 120patients have negative

family history. Out of 180 DR patients 30 had good glycemic control and 150 patients had poor glycemic control. Out of 180DR patients 100 patients were hypertensive (BP >145/95mmHg and 80 patients were normotensive (BP <145/90mmHg).

### SEROPREVALENCE OF AVIAN INFLUENZA IN COMMERCIAL POULTRY DURING WINTER SEASON AT KARACHI

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Avian Influenza has become endemic in poultry farming in Pakistan, being a potentially zoonotic disease, it remains a potential public health threat. For determination of seroprevalence of Avian Influenza in layers and broilers at Karachi, a total of 240 samples were collected from 30 flocks. From each flock 08 serum samples were taken during the months of November, December and January 2018-19. Serum antibody titers were determined by Hemagglutination inhibition (HI) test. HI test revealed that for H5, H7 and H9 subtypes, 32.08, 17.92 and 0% birds were seronegative, 43.75, 43.75 and 10% were exposed/immune and 24.17, 38.33. and 90% layer birds were infected. Further, in broiler birds, 72.1, 79.58 and 25.83% birds were seronegative, 27.9, 20.42 and 43.75% were exposed/immune and 0, 0 and 30.42 % were infected with H5, H7 and H9 subtypes respectively. The antibody titers against H5, H7 and H9 subtypes of Avian Influenza Virus in layer flocks were 2.13, 2.21 and 7.48 (log<sub>2</sub>), where as in broiler flocks titers were 0.7, 0.42 and 3.35 (log<sub>2</sub>), in November. In the month of December, antibody titers in Layers were 1.85, 2.23 and 7.15 (log<sub>2</sub>) while in broilers titers were 0.71, 0.375 and 3.30 (log<sub>2</sub>). The antibody titers in the month of January in Layers were 2.51, 4.13 and 7.10 (log<sub>2</sub>), while in broilers these were 0.18, 0.225 and 1.28 (log<sub>2</sub>), respectively. Seroprevalence data shows that Avian Influenza Virus infections are prevalent in both broiler and layer flocks however prevalence is higher in layers and infections with H9 subtype are more common.

### EPIDEMIOLOGICAL AND MOLECULAR STUDY OF *BABESIA BOVIS* IN DOMESTIC CATTLE OF DISTRICT BANNU KHYBER PAKHTUNKHWA

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Bovine babesiosis is a tick-borne disease of cattle caused by the protozoan parasites. In cattle mainly two species are involved in causing this disease i.e. *B. bovis* and *B. bigemina. Rhipicephalus (Boophilus* species) is main vector of this parasite. Pathological lesions include an enlarged pulpy spleen, a swollen liver, dark colored kidneys and anemia. The main objectives of this study were to determine the molecular detection of protozoan parasite (*Babesia bovis*) through Microscopy and PCR. A research work was carried out in district Banu. For these purpose nine different topographic areas were surveyed from May 2019 to August 2019. During this time span we have collected 100 blood samples from clinically suspected cattle of study area. The samples were preserved in refrigerator at 4c°. For detection of *B. bovis* Microscopy and PCR method was applied. Microscopic examination revealed that 67/100(67%) samples were positive and rest were found negative, while PCR study shows that 48/100(48%) samples were positive for *B. bovis*. These infections are reported predominantly in summer season. The research shows that prevalence ratio of this parasite is comparatively very high in study area and constitutes a major cause of economic losses at district Bannu. The reason behind this outbreak is due to lack of awareness regarding *B. bovis* and their vector (ticks).

#### EFFECT OF ALAN ON COMPLETE BLOOD COUNT PROFILE OF YOUNG FEMALE ADULTS

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Before the advent of electric lights about a century ago, humans were exposed to minimal light at night. At the end of the nineteenth century, with the invention of the electric light bulb, exposure to artificial light at night (ALAN) grew rapidly. Night shift work was introduced soon after. As technology boomed, humans encountered even more sources of light at night, including television and computer screens, smart phones and tablet computers. The increasing use of electric light has modified the temporal niche of humans, allowing them to be active at night. This change comes with obvious benefits, but we are also starting to see the dark side of night lighting. Recently, light pollution in the form of ALAN has been detected to cause multiple human cancers. ALAN can directly alter endocrine signaling from circadian dysregulation or disrupt melatonin production, or indirectly through inflammatory responses or elevated circulating stress hormones. The present study was aimed to investigate the effect of ALAN on complete blood count (CBC) profile in young female students. We found that the number of lymphocytes was significantly lower in ALAN group compared to control group. Moreover, HCT and MCH in ALAN Group were significantly lower as compared to control group. The other parameters (hemoglobin, red blood cells, white blood cells, neutrophils, monocytes, eosinophils, platelets, MCV and MCHC) remained unchanged. Taken together, the finding of present study demonstrated that ALAN could affect CBC profile of human.

# DIAGNOSTIC VALUE OF ANTI-TETANUS TOXOID ANTIBODY LEVELS IN HEMODIALYSIS PATIENTS

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Tetanus is a neuronal disorder which is characterized by severe muscle spasms caused by Clostridium tetani which releases the neurotoxin tetano-spasmin. The mortality rate of tetanus is very high i.e. 40% in adults and 90% in neonates. Tetanus vaccination being inexpensive is a standard in developed countries. It provides high level of protection and reduce tetanus incidence. Pakistani population is more at risk due to expanding populations, poverty and lack of education. The cold-chain defects, and inadequate facilities for transport of vaccines to target populations in remote areas is also a biggest factor of high risk population. Therefore, general levels of protection against tetanus are expected to be low. Patients with end stage renal disease and haemodialysis sequels in compromised immunity. Thus haemodialysis patients are likely to have even poorer tetanus toxoid antibody levels. The objectives of this study were to assess anti-tetanus toxoid antibody levels in haemodialysis patients and to assess immune response to single booster dose of tetanus toxoid vaccine. It was an expressive case series. Research was conducted at Pathology Department, Jinnah Hospital, Lahore. Duration of study was 9 months. At baseline, 36.5% patients were unprotected, 38.5% were partially-protected while 25.0% patients were fully-protected against tetanus. At follow-up, 46.5% were partially-protected while 53.5% patients were fully-protected against tetanus. When compared, this improvement in protection status from baseline was found statistically significant. It can be advocated from the results that in future practice, Hemodialysis patients should be routinely screened for anti-tetanus antibody status and booster dose should be given to partially protected patients to optimize the protection of patients against tetanus and unprotected patients should be referred for complete immunization.

#### SCREENING OF HUMAN BLOOD FOR DIFFERENT DISEASES ON MORPHOLOGY BASE

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The aim of our study was the screening of blood cells on the basis of morphology for different diseased. For this purpose, we randomly collect 318 blood samples from the students of university of swat. The collected samples were examined under the compound microscopic by using 100X standard method. During the observation 63 samples was normal and in remaining samples different types of morphological changes were observed. The percentage of morphological changes was 68.5%, in which Bite cell were 36%, Elliptocyte 34%, Tear drop cell 30%, Schistocyte 26%, Hypochromic cell 22.5%, Irregular contracted cell 16%, Echinocytes 15.5%, Roleaux 8%, Boat shape 6.5%, Sickle cell 5%, Keratocyte 4% and Acanthocytes 1.5%. In the analyzed slides, bite cell, elliptocyte, tear drop cell, schistocytes, hypochromic cell, irregular contracted cells were found frequently while echinocytes, boat shape cell, acanthocytes, sickle cells and keratocytes were found rarely. Morphogenetic characters are physical characters of an individual. In the present study different morphogenetic characters i.e. ear lobe attachment, clinodactyly (curved little finger) and tongue rolling were also observed. The result showed that individuals in which morphological changes were observed in the shape of cells, prevalence of these morphogenetic characters was observed as; 54% were able to tongue rolling, 31.8% were clinodactyly (curved little finger) and 22.2% were attach ear lobe.

#### FREQUENCY OF TYPHOID FEVER IN UPPER SWAT PAKISTAN

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Typhoid fever also called enteric fever, caused by Salmonella typhi. Salmonella typhi is 'negative bacteria and serotype of Salmonella enterica. Transmission of Salmonella typhi occurs due to contaminated of food and water, through fecal-oral- route. Typhoid fever is serious disease and health problem all over the world. Prevalence of Typhoid is mostly in Asia and developed countries. Five countries of Asia have high prevalence of Typhoid fever, Including Pakistan, India, Indonesia, China and Vietnam. Among these countries, prevalence is high in Pakistan and India, intermediate in Indonesia and low in China and Vietnam. Pakistan has more ratio as compare to India. Ratio of Typhoid fever in Pakistan is 451.7 per 100,000 populations per year, while ratio in India is 214.2 per 100,000 populations per year. Upper Swat is a district of Khyber Pakhtunkhwa Pakistan. The frequency of Typhoid fever was studied from August 2018 to June 2019 in Upper Swat. Upper Swat consists of three Tehsil, i.e Matta, Khwazakhela and Behrian. Total 270, 813 patients were visited to THO Matta in which 829 were Typhoid patients (0.306%), total 188,525 patients were visited to THQ Khwazakhela in which 1,856 were Typhoid patients (0.984%) and total 116,282 patients were visited to THQ Bahrain in which 1,942 were Typhoid patients (1.670%). So total 4,627/575,622 (0.803%) Typhoid patients were reported from August 2018 to June 2019 in upper Swat. Ratio of Typhoid fever in female are more as compare to male, Percentage in female is 52.62% while in male is 47.38%. Age wise, enteric fever is more in young's (having age 21-30 year i.e. 42.34%) as compare to old people (12.68%) and children (8.46%). Typhidot test and culturing are authentic technique for the determination of the Typhoid fever, while Widal test are used when time of infection is in the second week, In first weak infection are does not show through Widal test. The determination of frequency of Typhoid fever in upper Swat provide many information, about incident of Typhoid fever in this region, which can helps in future health policies.

# PREVALENCE OF UTERINE FIBROIDS (TUMORS) IN FEMALE PATIENTS FOUND IN CIVIL HOSPITAL HYDERABAD, LUMHS JAMSHORO, BHITTAI HOSPITAL HYDERABAD, ISRA UNIVERSITY HOSPITAL HYDERABAD

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Uterine fibroids (also known as leiomyomas or myomas) are the most common form of benign uterine tumors. Clinical presentations include abnormal bleeding, pelvic masses, Pelvic pain, infertility, bulk symptoms and obstetric complications. Current management strategies mainly involve surgical interventions, but the choice of treatment is guided by patient's age and desire to preserve fertility or avoid 'radical surgery such as hysterectomy. The managements of uterine fibroids also depend on the number, size and location of the fibroids. Other surgical and non-surgical approaches include myomectomy by hysteroscopy, myomectomy by laparoscopy, uterine artery embolization and interventions performed under radiologic or ultrasound guidance to induce thermal ablation of the uterine fibroids. There are only a few randomized trials comparing various therapies for fibroids. Further investigations are required as there is a lack of concrete evidence of effectiveness and areas of uncertainty surrounding correct management according to symptoms. The need for alternatives to surgical interventions is very real, especially for women seeking to preserve their fertility. There are many options which are proven to treat fibroids symptoms effectively.

# EFFECT OF *RAPHANUS RAPHANISTRUM* ON CHRONIC KIDNEY DISEASE INDUCE BY ETHANOL IN ANIMAL MODEL RATS

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Raphanus raphanistrum (radish) contain compound 1,1-diphenyl-2-picrylhydrazyl investigate recognized about antioxidant behavior. Many preclinical studies proposed that excessive alcohol intake has an effect on kidney. The aim of this study is to investigate the effect of Raphanus raphanistrum (radish) on Chronic kidney disease damage by reactive oxygen species or free radicals in animal model rats. Total of 18 rate were used in this study, divided into 3 groups and each group consist of 6 rats. Group 1 control (C), group 2 model (M) and group 3 test (T). Model and test group were treated with alcohol to produce chronic kidney disease by reactive oxygen spices for 9 weeks a dose of 1 ml. After that test group was treated with Raphanus raphanistrum juice for 4 weeks 80mg/kg body weight to determined it effect. Raphanus raphanistrum juice effect on behavior of rats through increases the locomotor activity and anxiety. The serum creatinine and uric acid level were significantly improved in T group. The reactive oxygen enzyme test shows that Super Oxide Dismutase (SOD) and Glutathione Peroxidase (GPx) was increase in T group. The Glutathione S-Transferases (GST) and Catalase (CAT) enzyme level was nearly same in C and T groups. This study concludes that compound 1,1-diphenyl-2-picrylhydrazyl found in Raphanus raphanistrum juice and possess strong antioxidant activity on Chronic kidney disease induce by ethanol through reactive oxygen species. There is need of more researches to determine the use of natural compound to treat different disease.

### EVALUATION OF EFFECT OF ANTI ASTHMATIC DRUGS AND ANTI-OXIDATIVE STATUS IN ASTHMATIC PATIENTS

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Asthma is a chronic lung disease that inflames and narrows the airways. Asthma causes recurring periods of wheezing, chest tightness, shortness of breath, and coughing. The inflammation in airways makes the airways swollen and very sensitive. The airways tend to react strongly to certain inhaled substances. When the airways react, the muscles around them tighten. This chain reaction can result in asthma symptoms. Symptoms can happen each time the airways are inflamed. Objective of the stud was to evaluate the effects of anti-asthmatic drugs and Anti-Oxidant status in Asthmatic Patients. The sample of sixty-five patients of asthma taking drug and sixty five patients of asthma who are not taking any drug were collected from Gulab Devi chest Hospital.5.0 ml sample was taken and subjected to centrifuge at 3000-4000 rpm for 10-15min for the separation of serum. MDA, AGEs, Catalase, SOD, Vitamins were estimated. The results of this study shows that the level of GSH was 0.13±0.03 in asthmatic patients while those who was not taking any drug was 0.17±0.04 it showed that it was statistically significant. The value of Vit C in drug taking patient was 0.44±0.11 and the other group those are not taking any drug 1.31±0.62 and it is also significant. The bilirubin level was 1.58±1.50 before treatment and the value was 0.75±0.48 after treatment and it is also significant. There is growing evidence that oxidative stress is key component involve in the pathophysiology of Asthma. The study shows that there is a significant effect on the NO, GSH, Vit C, Platelets, Bilirubin, SGOT, SGPT and Chloride of the asthmatic drug other factors are moderate or non-significant to the drugs.

#### CURRENT STATUS OF THEILERIOSIS IN CATTLE OF KHYBER PAKHTUNKHWA PAKISTAN WITH SPECIAL REFERENCE TO TAMS1 GENE

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Theileriosis a threat to farmers and livestock industry of KP Pakistan, being taking lives of cattles and other animals. Studying epidemiology and Sequence of Tams-1 gene to determine the phylogenetic position of *Theileria annulata* species of this region. Blood samples were randomly collected from 1800 cattles with a pro forma filled for each in order to calculate the position of *Theileria* infection from August 2015 to July 2016. The samples were screened using PCR. Primers for *Theileria parva* and *Theileria annulata* were used, only positive samples with *T. annulata* were detected no any sample was having *T. parva* infection. An overall prevalence of 12.78% (230/1800) by PCR was recorded in cattle, in which highest prevalence was in summer season. Gender wise high prevalence was found in female cattle, further calves were found more prone to the infection. Local breeds were found resistant as compare to the alien breeds. Tams-1 and 18S rRNA gene sequences of this region, showed relations with the sequences obtained from different countries. However it was inferred that both gene sequences are having a lot of sequence diversity, further these genes are not peculiar to a particular region.

#### PREVALENCE OF TOXOPLASMA GONDII IN CAT FAECES IN DISTRICT MARDAN

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Toxoplasma gondii is an obligate intracellular protozoan parasite with a worldwide prevalence, belongs to the phylum of the Apicomplexa. The present epidemiologic survey study investigated the record of prevalence of T.gondii from felis cat feces in district Mardan and district Sawabi, Khyber Pakhtunkhwa (KPK). Total number of fecal samples observed is 250. These samples were examined under compound microscope by using wet mount method. All the collected samples were brought to the Abdul Wali Khan University Mardan (AWKUM), zoology department, parasitology lab of microscopy. Overall of 250 fecal samples, 47 were positive showing 18.8% and 203 were negative showing 81.2%. These samples were positive for T.gondii oocyst. Total samples collection took 3 months (Feb 2017, March, April 2017) from Pohan colony 66 samples were collected, 20 of which are collected in Feb, 28 in March and 15 in April. In these 66 samples 14 samples were positive showing 25.2% prevalence. In these three months 250 samples were collected respectively, from Ghaznavi colony Mardan, out of 39 samples only one sample was positive showing 15.6% prevalence. From Shankar area 8 samples were collected in February which are all negative (have no oocyst of T.gondii) from Baghdada Mardan total 48 samples total 48 samples were collected ,26 in Mardan, 15 in march ,7 in April in which 14 samples were positive showing 19.2% prevalence. From district Sawabi 20 samples were collected.6 in February 0 in March and 14 in April in which 5 were positive and 15 were negative showing 8% prevalence. From Qasmi tehsil Katlang 22 samples were collected in February and march respectively, out of these 27 samples were positive showing 10.8 prevalence. The recent study provides the percentage 18.2% of cat fecal samples testing positive for *T.gondii* oocysts in district Mardan. According to Lilly the percent positive in this study (6%) was significantly higher than expected when compared with 10 of the samples, comparable with 2, and less than expected when compared with the feral cat population in Ethiopia.

### 4. MICROBIOLOGY

### STUDY OF ANTIMICROBIAL POTENTIAL AND ANTIBIOTIC SUSCEPTIBILITY OF BACTERIA ISOLATED FROM SOIL AND WATER OF RIVER RAVI

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Microbiota has been proved to be a natural source of chemical compounds which have shown great diversity. Many microbes produce secondary metabolites which are very important bioactive compounds. The bioactive compounds are the origin of good number of marketed antibiotics. These compounds have been used in pharmaceutical and drug industry. These compounds have unique chemical nature and structures. Exploration of soil and water reservoirs is required which are major source of microbes. Present study is designed to explore microbes from soil and water brought in by River Ravi Lahore, Punjab. Two samples each of soil and water have been collected from different sites of River Ravi. A total of about 50 isolates were checked for their antimicrobial activity against Klebsiella pneumoniae, Escherichia coli, Bacillus subtilis, Bacillus licheniformis, Pseudomonas aeruginosa and Salmonella sp. Preliminary screening by cross streak method showed 19 potent strains. Agar well diffusion method was used to check the antimicrobial potential of bioactive compounds of obtained 19 isolates. Secondary screening confirmed 03 potential isolates.

#### ANTIMICROBIAL AND ANTI-LARVICIDAL ACTIVITY OF TERRESTRIAL STREPTOMYCES

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Microbial diseases are continuously increasing every year and becoming big global threat to human and wildlife health. There are more than 300 known diseases transmitted by bacteria, fungi, viruses and mosquitoes etc. So, search for new novel bioactive compounds from actinomycetes to control these pathogens and their resistance mechanisms is the need. 14 actinobacterial strains were isolated from six samples. These strains were subjected to morphological, biochemical and differential media characterization. The antibiotic susceptibility test of all the isolated strain was done using disc diffusion method. The actinobacterial strains AB1, AB7, GB1, GB7 and GB8 showed resistance against 04 antibiotics (ampicillin, rifamycin, lincomycin and erythromycin) and were subjected to screen for potent antimicrobial compounds following primary and secondary screening methods. The preliminary screening was done against E. coli, P. aeruginosa, K. pneumonia, B. subtilis and B. licheniformis using standard cross streak method. Ethyl acetate extraction was performed in order to isolate bioactive compounds from potent 05 strains. The ethyl acetate crude extracts were dissolved in Dimethyl sulfoxide (DMSO) and used to perform secondary screening and larvicidal activity. In primary screening, actinobacterial strain AB7 showed highest zone of inhibition (ZI) (24 and 27 mm) against B. subtilis and B. licheniformis respectively. Actinobacterial strain GB8 and GB7 showed highest ZI (19 mm) against E. coli. All the isolated strains showed relatively small ZI against K. pneumonia and P. aeruginosa. Using agar well diffusion method, it was observed that actinobacterial strain GB6 showed highest ZI (14 mm) against P. aeruginosa. Other strains (AB1, AB7, GB7 and GB8) also showed excellent zone of inhibition (12-10 mm) against P. aeruginosa. The ZI against E. coli, B. subtilis and B. licheniformis was in the range of 6-11 mm. The minimum inhibitory concentration (MIC) of these 05 actinobacterial strains was in the range of 1.3 to 3.5 mgmL<sup>-1</sup> and minimum bactericidal concentration (MBC) was in the range of 1.9 to 4 mgmL<sup>-1</sup> against the selected pathogens. The actinobacterial strain AB7, AB1 and GB6 showed 100% mortality and GB7 and GB8 showed 83% and 68% mortality at 1000ppm against Anopheles 3rd instar larvae. These findings indicated that actinobacterial isolates have the ability to produce bioactive compounds with potent activity against Gram positive and negative pathogenic bacteria and have the ability to kill mosquitoes. This could be a good source of novel antibiotics and natural insecticides.

# ISOLATION AND CHARACTERIZATION OF PARTIALLY PURIFIED CELLULASES FROM GUT BACTERIA OF *ODONTOTERMES OBESUS* RAMBUR (BLATTODEA: TERMITIDAE)

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The rising alarm about the insufficiency of fossil fuels, the emission of greenhouse gasses and air pollution by incomplete combustion of fossil fuels, biomass conversion and other industrial process have resulted in an increasing focus on the use of cellulases. The present study was undertaken to isolate and characterize the cellulases from gut bacteria of a higher termite 'Odontotermes obesus' collected from different mounds located at Quaid-i-Azam University, Islamabad, Pakistan. The cellulase producing bacteria from termite gut were identified by morphological, biochemical and molecular analysis. The partial purification of cellulases was carried out by ammonium phosphate precipitation and dialysis. Seven different bacterial strains (O-01, O-02, O-03, O-04, O-13, O-14, and O-15) were isolated and screened by CMC ager media and Congo red assay. The specific activity of crude and precipitated enzyme (cellulase) from seven strains was 0.55, 0.59, 0.34, 0.48, 0.42, 0.56, 0.35 and 0.74, 0.83, 0.30, 0.38, 0.45, 0.70, 0.31 U/mg respectively. The agro-waste material as a carbon source for cellulase production by bacterial strains was studied with maize bran, sugarcane baggase, wheat bran, and rice bran. The bacterial isolates O-01, O-02, O-03 and O-13 exhibit highest activity towards sugarcane followed by rice bran and wheat bran respectively while for O-04 and O-15 maximum cellulase production was reported for sugarcane barn. The SDS-Page indicated presence of exo, endo-β-1, 4- glucanases and β- glucosidase on basis of molecular weight was 53, 55, 70 and 80, 50, 68kDa. Cellulase enzyme activity was characterized with optimum temperature, pH, divalent ions, organic solvents, inhibitors and surface active agents. The optimum temperature turned out to be 35°C for O-04, O-15 strains and 45°C for O-01, O-02, O-03 and O-14. The optimum pH was determined 8 for strains O-013 and O-14 similarly pH-7 for strains O-02, O-13, O-14 and pH 7.5 for strain O-04respectively. The cellulases activity was stable in range of 6.8-10 for strain O-13. The cellulases activity was enhanced in the presence of organic solvents like Methanol, Toluene, Formaldehyde and DMSO. The cellulase activity was stimulated by metallic ions like Ca, Mg and Na whereas strongly inhibited by Hg and Ag. Cellulases from all strain are stable toward most of organic solvent only Benzene inhibited their activity. All strains of cellulases are quite stable in the presence of surfactants, H<sub>2</sub>O<sub>2</sub> and NaClO enhanced while EDTA slightly inhibits the activity. Kinetic analysis of purified enzyme indicated the Km and Vmax to be 2.24 mg ml<sup>-1</sup> and 4.05 µg ml<sup>-1</sup>min<sup>1</sup>, respectively. High activity profile and resistant to different detergent, metals and organic solvent make the cellulases desirable in different industrial applications.

### IDENTIFICATION AND MOLECULAR CHARACTERIZATION OF OIL DEGRADING BACTERIA ASSOCIATED WITH FISH OF INDIAN OCEAN

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Biodegradation of oil by microorganisms is the ultimate solution for the removal of marine oil pollutants. In this

study we identified and characterized fish associated oil degrading bacteria. A total of 11 fish samples were collected from Indian ocean, near oil port at Karachi. Surfaces, gills and guts of fish were used for bacterial isolation. Millions of bacteria g<sup>-1</sup> of gills and guts and cm<sup>-1</sup> of surface were found associated with fish. Selection of potential isolates were carried out using Nutrient Agar (NA) and MSA media with gradual increase in crude oil concentration (0.2%, 0.5%, 0.7%, 1%, 2% and 5% v/v). Finally, 4 isolates out of 36, were selected for further identification. Two of the isolates (G4A and G3A) were Gram positive while two (G2A and L2C) were Gram-negative rod-shaped bacteria. Characterization based on their 16S rRNA gene sequencing, showed that isolates were belonging to *Bacillus velezensis* (G4A), *Bacillus flexus* (G3A), *Pseudomonas brenneri* (G2A) and *Pseudomonas azotoforman* (L2C). Phylogenetic analysis showed that these bacterial species are adapted to high concentration of oil pollution and can degrade oil in the marine environments. These bacterial species can be exploited for the removal of oil pollutants from aquatic environment.

### ISOLATION AND MOLECULAR CHARACTERIZATION OF *CLOSTRIDIUM PERFRINGENS*IN THE SMALL RUMINANTS OF DISTRICT KOHAT

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Clostridium perfringens is a gram-positive pathogenic bacterium, causing enterotoxemia and gastrointestinal (GI) tract diseases in domestic animals. C. Perfringens type D toxinotype is the most common in livestock, liable for inducing enterotoxemia. In this study, we have collected total 206 samples of diseased small ruminants (sheep and goats) from eight different topographic areas of district Kohat. A total of 94 goat's samples were observe for C. perfringens using Thioglycollate broth. Result showed that 42 goat's samples and 46 sheep samples stood identified up for C. perfringens. Same as agar, 46 sheep samples while 42 goat's samples stood up for C. perfringens by Gram staining. After Gelatin liquefaction test, 37 sheep samples out of 112 were identified up for C. perfringens, while 23 goat's samples out of 94 were identified up for C. perfringens. The percentage of positive cases was 37 (33.04%) out of 112 in sheep and 23 (24.5%) out of 94 in goats for C. perfringens based on isolation and PCR amplification of Alpha toxin gene. The overall prevalence was 57.54% for C. perfringens present in rams and kids of district Kohat. Our results concluded that C. perfringens type D is highly predominant in domestic small ruminants of district Kohat.

### DIAGNOSIS OF MYCOBACTERIUM BOVIS THROUGH CONVENTIONAL AND MOLECULAR TECHNIQUES

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- A cross sectional study was carried out in order to establish a rapid and stable method for diagnosis of Mycobacterium tuberculosis infection through conventional and Molecular techniques and decrease the side effects of late diagnosis of patients and health system. Decrease long process of laboratory identification of the infection rate of *Mycobacterium bovis* and establishing new diagnosis methods was one of the reasons for this study. To accomplish this objective, collected milk and blood specimens from 100 BTB patients with clinical doubt of pulmonary tuberculosis were studied with both molecular and traditional techniques, Polymerase Chain Reaction (PCR), Enzyme-linked immunosorbent assay (Elisa) (IgG and IgM), Acid-Fast Bacillus stain (AFB) and culture methods were compared. The

sensitivity and specificity of all methods were determined by using the PCR results as the gold standard. The overall sensitivity, specificity, positive predictive value and negative predictive value of AFB were 47.37, 49.38, 18.0 and 80.0%. These values for culture method was 66.67, 52.94, 20.0 and 90.0% and for IgG antibody were 70.0, 55.0, 28.0 and 88.0%, IgM antibody were 76.47, 55.42, 26.0 and 92.0% and for PCR were 100.0, 60.97, 36.0 and 100.0% respectively. It was concluded that maximum sensitivity and specificity can be achieved by PCR method.

# COMPARISON OF ANTIBACTERIAL EFFECTS OF PURE ALOE VERA AND DOXYCYCLINE AGAINST *STAPHYLOCOCCUS AUREUS* AND *STREPTOCOCCUS AGALACTIAE* ISOLATED FROM MASTITIC MILK OF BUFFALO

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Aloe vera, also called a miracle plant because of its broad spectrum therapeutic use against many diseases caused by bacteria, fungi, virus and other parasites. Hence, present experiment was conducted to examine the antibacterial effect of pure Aloe vera and compared with doxycycline. During the study, total 52 milk samples (quarters) were collected under aseptic conditions from the dairy farms in the vicinity of Tandojam. Samples were brought in the Department of Veterinary Pharmacology for further process. After isolation and identification, the organisms were tested against different concentrations of pure Aloe vera and doxycycline to determine their minimum inhibitory concentration (MIC). For this purpose, similar concentrations of pure Aloe vera and doxycycline were used, i-e, C1= (30), C2= (15), C3= (7.5), C4= (3.75), C5= (1.87), C6= (0.93), C7= (0.46), C8= (0.23), C9= (0.11), C10= (0.05), and C11= (0.02) μg/μl were used (MIC). The MIC was evaluated on the basis of turbidity and transparency of the medium. Out of 52 milk sample (quarters), 30 quarters (57.69%) were found positive, 22 quarters (42.30%) were found negative. Further analysis of positive quarters indicated that S. aureus and S. agalactiae pure was isolated from 17 quarters (56.66%) and from 9 quarters (30%), respectively. The colonies of the both bacteria were isolated from 4 quarters (13.33%). The MIC for pure Aloe vera against S. aureus and S. agalactiae were determined as (3.75 μg/μl) and (1.87 μg/μl), respectively. Whereas, As compared to pure Aloe vera, Doxycycline was found most effective, the MIC against S. aureus and S. agalactiae were determined as (0.23 µg/µl) and (0.46 μg/μl), respectively. Analysis of variance showed significant difference between treatments and their various concentrations used to evaluate MIC. It is concluded that both treatments produced susceptibility against isolated organisms. As antibiotic resistance is rapidly developing in microorganisms due to prolonged use of conventional medicine, therefore, it is suggested that Aloe vera can be a good alternative against mastitis causing organisms.

# PREVALENCE AND ANTIMICROBIAL SUSCEPTIBILITY OF FOODBORNE PATHOGENIC BACTERIA IN RAW FISH SAMPLES COLLECTED FROM PONDS AND RETAIL MARKET OF DADU, SINDH, PAKISTAN

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Research has declared that fish and its products are the major and cheap source of animal origin protein after meat and chicken, however, it is being recognized as a major source of transmission for foodborne diseases. A total of 120 samples from three different fish species *i.e.*, Labeo rohita (LR), Catla catla (CC) and Cirrhinus mrigala (CM) were collected and processed for isolation of foodborne pathogens using conventional culture technique. The samples were collected from two different sources *i.e.*, ponds and fish retail market of district Dadu, Sindh, Pakistan. According to results, E.coli, Salmonella spp., Shigella flexneri, Pseudomonas flavescens, Pseudomonas aeruginosa,

Staphylococcus aureus, Streptococcus agalactiae, Klebsiella pneumoniae, Staphylococcus epidermidis, Klebsiella oxytoca and Streptococcus pyogenes were isolated from both ponds and market fish samples. Prevalence of foodborne bacterial pathogens were high in market samples as compared to pond samples. Results of antimicrobial susceptibility profile carried out by Clinical and Laboratory Standards Institute (CLSI) method showed that all bacterial isolates were resistant to four or more antimicrobials thus regarded as multidrug resistant. There was no difference in contamination level between LR, CC and CM. These results demonstrated that raw fishes in district Dadu contained high level of multidrug resistant foodborne bacterial pathogens.

# EFFECTS OF PROBIOTIC SUPPLEMENTATION ON GROWTH AND CONFORMATION OF GOAT KIDS

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An experiment was carried out at the Department of Livestock Management, Faculty of Animal Husbandry and Veterinary Sciences, Sindh Agriculture University Tandojam to compare the growth, body conformation and economics of non-descriptive male goat kids under different management systems. Twelve kids about 6 months of age were divided into two groups and kept under different management systems and fed with a ration supplemented with probiotics. The group A kids were raised in a semi-intensive management system, allowed to graze and fed with concentrate ration mixed with commercially available probiotics. While, the kids in group B were subjected to an intensive management system restricted to graze and received a concentrated ration mixed with probiotics and green forage at the animal shed. The total duration of the experiment was eight weeks and the observations on the growth and conformation features (girth, height and length) were recorded on weekly basis. The results revealed that the average final body weight of group A kids were (15.45 kg) significantly (P<0.05) higher than that of kids in group B (13.86 kg). In addition, the body conformation i.e. girth, height and length (61.33 cm, 53.66 cm and 44.5 cm respectively) of kids in group A were also observed significantly higher (P< 0.05) as compare to the kids of group B (58.83 cm, 52 cm and 43.33 cm respectively). While, comparing the economic aspects of kids of experimental groups, it was observed that group A earned more net profit (2338 rupees / kids) than group B (1491 rupees / kid). In conclusion more appropriate performance in terms of body weight and body conformation traits were recorded in goat kids raised under semi-intensive management system and fed concentrated ration mixed with probiotics supplements, and were found more profitable.

### DISTRIBUTION AND ANTIMICROBIAL SUSCEPTIBILITY PATTERN OF BACTERIAL PATHOGENS CAUSING URINARY TRACT INFECTIONS AMONG THE POPULATION OF ISLAMABAD, PAKISTAN

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The aim of the present study was to investigate distribution and antimicrobial susceptibility pattern of bacterial pathogens isolated from patients having urinary tract infections (UTIs). A total of 600 urine samples were collected from three hospitals of Islamabad namely, Federal Government Services Hospital, Pakistan Institute of Medical Sciences and Capital Development Authority Hospital. Samples were analyzed to evaluate the prevalence of UTIs among the population. Urine samples with UTIs were cultured over night on Cystine Lactose Electrolyte Deficient agar. The causative agents of UTIs were determined by gram staining. Antimicrobial susceptibility of uropathogens was evaluated by Kirby Baur disc-diffusion method. Among 600 samples, 52% showed a significant bacteriuria with a disease predominance in females (80.76%) over males (19.23%; P < 0.0001). Females within the age groups 15-40

years and >65 years and elderly males of >65 years showed high prevalence of UTIs. Gram negative bacteria (89.74%) were more prevalent with *Escherichia coli*. To determine antimicrobial susceptibility, a total of 13 antibiotics were tested namely, ampicillin, amoxicillin-clavulanic acid, cefalothin, cefuroxime, ceftazidime, meropenem, gentamicin, amikacin, ciprofloxacin, nalidixic acid, trimethoprim-sulfamethoxazole, chloramphenicol and nitrofurantoin. Among these Ampicillin showed the least sensitivity whereas Meropenem was the most effective drug against all isolated uropathogens. It is concluded that uropathogens have developed a high resistance against most of the tested antibiotics.

### COMPARATIVE EFFICACY OF ALOE VERA AND LEVOFLOXACIN AGAINST CORYNEBACTERIUM PYOGENES AND STAPHYLOCOCCUS EPIDERMIDIS ISOLATED FROM CAMEL WOUND.

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The present study was conducted to determine the minimum inhibitory concentration (MIC) of pure Aloe vera and compared with commonly used antibiotic, Levofloxacin against Staphylococcus epidermidis and Corynebacterium pyogenes isolated from camel wound. During this study, 70 wound samples were collected, from veterinary clinics and nomadic animals in the vicinity of Hyderabad and Tando Allah Yar and were brought to the Department of Veterinary Pharmacology, Faculty of A. H. and Vet. Sciences, SAU Tandojam for further process including isolation and identification of organisms. To record MIC against isolated organisms, different concentrations of pure Aloe vera and Levofloxacin were used i.e  $20\mu g/\mu l$ ,  $10\mu g/\mu l$ ,  $5\mu g/\mu l$ ,  $2.5\mu g/\mu l$ ,  $1.25\mu g/\mu l$ ,  $0.62\mu g/\mu l$ ,  $0.31\mu g/\mu l$ ,  $0.15\mu g/\mu l$ ,  $0.07\mu g/\mu l$ ,  $0.03\mu g/\mu l$  and  $0.01\mu g/\mu l$ . The MIC of the isolates was determined by turbidity and translucency of the cultured medium. Results of this study revealed that out of 70 wound samples, 50 were found positive. Among those 29(58%) and 21(42%) were found positive for Staphylococcus epidermidis and Corynebacterium pyogenes respectively. It was found that Staphylococcus epidermidis stopped growth at 5µg/µl and 0.31 µg/µl of pure Aloe vera and Levofloxacin respectively. The mean susceptibility value of Staphylococcus epidermidis was noticed 7.77 μg/μl for pure Aloe vera and 0.55 μg/μl for Levofloxacin. Whereas, the highest and lowest concentration at which the growth of Staphylococcus epidermidis inhibited was 10 and 0.62µg/µl and 5, 0.31µg/µl observed for pure Aloe vera and Levofloxacin respectively. Whereas Corynebacterium pyogenes stopped its growth at 5 µg/µl and 0.15 µg/µl of pure Aloe vera and Levofloxacin respectively. The mean MIC value of Corynebacterium pyogenes was observed as 7.77 µg/µl for pure Aloe vera and 0.25 µg/µl for Levofloxacin. Whereas, the highest and lowest concentration at which the growth of Corynebacterium pyogenes inhibited was 10 and  $0.31\mu g/\mu l$  and 5,  $0.15 \mu g/\mu l$  observed for pure Aloe vera, and Levofloxacin respectively. The results revealed that pure Aloe vera extract has considerable antibacterial activity and supports the traditional use of plants so that they can be used as an alternative to antibiotics in order to reduce the emergence of antibiotic resistance.

### PREVALENCE, RESISTANCE PROFILING AND RISK FACTORS ANALYSIS OF UROPATHOGENIC ESCHERICHIA COLI

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Escherichia coli (E. coli) are very well known to cause urinary tract infections. The most common dilemma today we are facing is the decreased level of protection from infections by antibiotics. To determine its prevalence, antibiotic resistance profiling and associated risk factors, urine samples were processed by routine staining techniques, biochemical tests followed by culture techniques using disc diffusion method. A total of 750 urine samples were collected from City Lab and Diagnostic Centre, Lahore. Total positive isolates were 330, out of which 142 (43 percent) were E. coli. The prevalence of E. coli in males is 44 (30.9 percent) and in females 98 (69.0

percent). *E. coli* were found highly sensitive to Meropenem (99.9 percent), Imipenem 138 (97.1 percent), Amikacin (94.3 percent), Tazobactum-piperacillin 117 (82.3 percent). The sensitivity pattern to other antibiotics was Cefoperazone/ sulbactam 116 (81.6 percent), Fosfomycin 98 (69.0 percent), Gentamycin 92 (64.7 percent), Aztreonam 46 (32.3), Cefuroxime 45 (31.6 percent), Nitrofurantoin 44 (30.9 percent), Ciprofloxacin 43 (30.2 percent). Risk factors associated with this study are kidney stones, urinary catheters and diabetes. The present study data may help in providing the guideline to the clinicians for suggesting appropriate antibiotics in treating *E. coli* UTIs. In future there should be further studies on epidemiology and Genetic causes of antibiotic resistance in uropathogenic *E. coli* in various regions of Pakistan.

### MOLECULAR CHARACTERIZATION OF NEW DELHI METALLO-BETA-LACTAMASE-1 GENE FROM CLINICAL ISOLATES OF *ESCHERICHIA COLI* FROM TERTIARY CARE HOSPITALS LAHORE, PAKISTAN

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The development of antibiotic resistance has become a global phenomenon. The evolving status of antibiotic resistant bacteria especially extended spectrum β-lactamase and carbapenemase-producing members of Enterobacteriaceae poses a serious health concern all over the world. Present study was designed to determine the antibiotic susceptibility pattern and genetic characterization of carbapenem resistance in Escherichia coli that was isolated from various tertiary care hospitals of Lahore. E.coli (n=90) from different clinical samples (120) including pus, blood and urine was identified by various biochemical testing. All isolated samples were subjected to antibiotic susceptibility testing by disc diffusion assay, for the detection of carbapenem resistance. The phenotypically Modified Hodge Test was also performed on carbapenem resistant E.coli. Among all the tested antibiotic disks E. coli was highly resistant to imipenem (85.6%), meropenem (88.9%), ertapenem (87.8%), ampicillin (90%), amikacin (80%), tobramycin (91.1%), tigecycline (20%), tazobactam (26.7%) and aztreonam (66.7%). Among gender, the carbapenem resistance was more in male (82%) than female (80%). Among various specimens, the carbapenem resistance in blood samples was 77% with respect to other samples like urine (75%), body fluids (70%) and pus (63%). The carbapenem resistance is attributed to New Delhi Metallo β-Lactamase-1 gene (blaNDM-1). So, carbapenem resistant gene was amplified by polymerase chain reaction. Molecular characterization of these isolates revealed that 20 isolates carried the carbapenem resistance gene blaNDM-1. It is concluded that carbapenem resistance is on the rise in hospitals set up and rapid tests for detection of antibiotics resistance should be established and strict monitoring with rational use of antibiotics is required. Furthermore, it is suggested that use of lactam ring containing antibiotics may be closely monitored in clinical and veterinary set up to delay the development of resistance against it.

# CONTRIBUTION OF PHOSPHOROUS SOLUBILIZING *ALOE VERA* RHIZOMICROBIOME IN WHEAT GROWTH PROMOTION

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The global population hike entails the need for uplifting agricultural productivity with significant decrease in the use of synthetic chemical fertilizers and pesticides. In this study, twenty plant growth promoting rhizobacteria (PGPR) were isolated from *Aloe vera* rhizosphere. Bacterial isolates were morphologically and biochemically characterized by quick test (QTS 24) bacterial identification kits. Three strains were identified as species of *Klebsiella*, two as *Enterobacter* and other isolates as species of *Proteus* and *Bacillus*. Variable production of extracellular hydrolytic enzymes including protease, cellulase, amylase, and lipase was observed by all strains. All

strains were screened for phytohormones indole-3-acetic acid and production of ammonia. Five strains i.e., AURS1 (*Klebsiella oxytoca*), AURS3 (*Klebsiella* sp.), AURS5 (*Enterobacter cloacae*), AURS10 (*Enterobacter* sp.) and AURS19 (*Klebsiella* sp.) were able to solubilize insoluble phosphate and potassium (solubilization index >=1.2-1.6cm) and selected as bio-inoculums of wheat. Wheat seedlings treated with phosphate solubilizing strains significantly increased germination rate and seedling vigour (VI) as compared to un-inoculated control plants. Wheat plant experiments were carried out in two experiment sets; A (without fertilizer) and B (with fertilizer) in a growth chamber in RCBD. Upon harvesting, root and shoot lengths, fresh and dry root and shoot weights were noted. For experiment set A, AURS5 (*Enterobacter cloacae*) showed maximum root lengths, dry root and shoot weights whereas AURS3 (*Klebsiella* sp.) showed maximum shoot lengths without fertilizer. In case of experiment set B, AURS5 (*Enterobacter cloacae*) showed maximum root lengths and dry shoot weights whereas AURS10 (*Enterobacter* sp.) showed maximum dry root weights and shoot lengths with fertilizer. The results indicated that phosphate solubilizing PGPR can be eco-friendly and cost effective candidates to improve the phosphorus uptake by plant resulting in increased plant growth and yield.

# COMPUTATIONAL AND MOLECULAR CHARACTERIZATION OF NDM-1 METALLO-B LACTAMASES RESPONSIBLE FOR MULTIDRUG RESISTANCE IN BACTERIA

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New Delhi Metallo-β-lactamases (NDM-1) is monomeric and plasmid- encoded enzymes that have become widely concern for its biological activities to catalyse the hydrolysis of mostly β-lactam. Due to the presence of this enzymes bacterial resistance take place mostly all β-lactam antibiotics and reported in *Klebsiella pneumonia*, *Escherichia coli* and *Acentobacter baumanii*. A serious concern towards the successfully treatment of patients in clinics are multi drug resistance of nosocomial pathogens. B-lactamases are ineffective against these superbugs and it is very necessary to make novel drugs with selective potential for the treatment of the diseases. In present study Extensive data mining, Molecular docking and Energy minimization were performed to select the best inhibitors compounds and NDM-1 enzyme showed lowest energy (-7.4 Kcal/mol) with *Lenapenam* as potential inhibitor for antibacterial substances with the help of molecular docking. Hence, it is concluded that these potential inhibitors may be used to treat the infectious diseases by overcoming the antibiotics resistance.

### CHARACTERIZATION OF PSEUDOMONAS AERUGINOSA AND IDENTIFICATION OF ITS GENETIC VARIANTS USING COMPUTATIONAL APPROACH

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Pseudomonas aeruginosa is a rod-shaped, motile, Gram-negative pathogen that causes acute and chronic nosocomial infections such as bacteremia, pneumonia, progressive cystic fibrosis, and urinary tract infections. Excessive use of antibiotics and emerging resistance of P. aeruginosa against an array of antibiotics has led to the development of several multidrug-resistant strains. Moreover, biofilm formation ability of this species adds to its resistance against antimicrobial agents. Following study evaluated the antibiotic resistant strain of P. aeruginosa, its biochemical characterization, and analysis of crystal structures of virulence proteins. Comparative analyses on conserved virulence domains and its most common relatives through phylogeny were also evaluated. Biochemical characterization was based on indole-3-acetic acid production, methyl red and phenol red bioassays, and motility tests. Furthermore, in-silico approach was used for understanding homology and evolutionary relationships through phylogenetic analysis of P. aeruginosa and its genetic variants through CLUSTALW and Phylogenyfr. Moreover, the functional proteins, conserved domains and crystal structures were analyzed by using PDB software.

### IDENTIFICATION AND GENOMIC CHARACTERIZATION OF BACTERIOPHAGE AGAINST PSEUDOMONAS AERUGINOSA

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Pseudomonas aeruginosa (P. aeruginosa) is a pathogenic opportunistic bacteria and one of the leading cause of nosocomial infections. Due to an increase antibiotic resistance among P. aeruginosa, there is need for an alternative treatment to cure infections caused by Multidrug-Resistant (MDR) bacteria and bacteriophage could be used for this purpose. Previously isolated novel phage which was named as JHPS against P. aeruginosa carries remarkable lytic ability towards clinical isolates of P. aeruginosa on double layer agar technique. The main aim was to identify effectiveness of bacteriophage against interspecies and intraspecies multidrug resistance bacteria (Acinetobacter sp. Pseudomonas sp. Escherichia coli and MRSA) with spot test method. The results showed that the JHPS phage is very host specific. All these characteristics demonstrated that JHPS phage is a novel candidate having a strong lytic potential for MDR P. aeruginosa strains. However, biochemically identified JHPS phage against P. aeruginosa was further characterized on the genomic basis by extraction of bacterial DNA followed by Polymerase chain reaction. For phage morphological analysis Scanning electron microscopy and Transmission electron microscopy was performed to classify the phage according to (ICTV) which revealed the characteristic features of the Tectivirdae family with absence of contractile tail, and with linear dsDNA, and protein rich internal membrane, which is enclosed in an icosahedral head structure. This study may provide the basis for a very promising candidate for phage therapy. JHPS phage could be considered as a useful candidate to control P. aeruginosa infections.

# IDENTIFICATION OF BACTERIAL ISOLATES ON CULTURE OF CEREBROSPINAL FLUID IN CHILDREN PATIENTS SUSPECTED WITH MENINGITIS

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Bacterial meningitis is a severe contagious disease of children and adults, and can be fatal in all age groups. In recent years, due to the development of polysaccharide and conjugate vaccines, its incidence has been decreased. However, 1.2 million cases of bacterial meningitis are still reported each year. Bacterial meningitis occurrence and mortality number vary depending on the geographic region, pathogen type, and age group. The present study was conducted to identify common pathogens and their antimicrobial sensitivity in cerebrospinal fluid (CSF) that causes meningitis in children. It was an observational and descriptive study. The study included 200 CSF samples of children suspected with meningitis. Each sample was processed for microscopic examination, culture and antimicrobial sensitivity. All samples were negative for the Ziehl-Neelsen stain ruling out the possibility of mycobacterial infection. Of these 18 samples (9.0%) were positive for gram stain, out of which 14 (78%) were gram negative and 04 (22%) were gram positive. Gram negative isolates after culturing showed the presence of Acinetobacter baumannii (27.8%), Escheria coli (11.1%), Serratia marcescence (11.1%), Enterobacter cloacae (11.1%), Haemophilus influenza (11.1%), and Psuedomonas spp. (5.55%). The gram positive bacteria included Streptococcus pneumoniae (5.55%), Methcilin resistant Staphylococcus aureus (5.55 %), Methcilin sensitive Staphylococcus aureus (5.55%), and Entrococcus faecalis (5.55%). Gram negative and gram positive bacteria depicted variable antibiotic suceptibility. To prevent permanent neurological sequelae, apt adoption of prompt diagnosis and treatment is important.

# EPIDEMIOLOGY OF METHICILLIN RESISTANT STAPHYLOCOCCUS AUREUS (MRSA) IN DIABETIC PATIENTS

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Diabetic Foot Infection (DFI) is common amongst diabetic patients in Pakistan. Many cases of Methacilin Resistant Staphylococcus aureus (MRSA) have been surfaced in the last decades in Pakistan. The purpose of this study was to check the prevalence of MRSA infection and to determine the most common drug resistance against S. aureus in diabetic patients. Data of diabetic patients was collected from Sheikh Zayed Hospital. A retroactive examination of total 175 samples were studied during December 2017 to May 2018 and screened for MRSA using mainly cefoxtin blood agar and Oxacillin Resistant Screening Base Agar (ORSBA). Specimens were cultured considering the microbiological techniques which are recommended by Clinical and Laboratory Standards Institute (CLSI) along with performance of antibiotic susceptibility tests. Disc diffusion point assay was used to measure sensitivity of isolated petients while to find out the Minimum Inhibitory Concentration (MIC) of various antibiotics, broth dilution process was performed. İsolates were also tested against commonly used antiboitics. The colonies were mostly found in the foot site. To isolate MRSA, ORSBA was prefereable over the other agar medium. Major risk issues for colonization of MRSA were male patients. Isolated patients were responsive to vancomycin and linezolid. Most of the isolates were also responsive to rifampicin. Among all the pathogens discovered, S. aureus dominates in patients suffering from diabetics. It is considered that the prevalence of methicillin resistant S. aureus (MRSA) is 15-30% in diabetic infectinos mainly targeting foot wounds and skin ulcers. High level of resistance was shown by penicillin and ampicillin. Meausres like beter hygiene and proper use of antibioites should be adopted to cure the infection.

### SCREENING AND *IN-SILICO* ANALYSIS OF PLANT GROWTH PROMOTING AND MERCURY RESISTANT RHIZOBACTERIAL STRAINS

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Pollution of heavy metals such as mercury (Hg) in the rhizosphere poses serious danger to plant growth and yield and then ultimately human health. In the present study, a total of 30 bacterial isolates were qualitatively screened out on the basis of resistance against mercury at the concentration of 30 µg/ml of HgCl<sub>2</sub> and growth on selective medium i.e. nitrogen free mannitol (NFM) for nitrogen fixing bacteria (NFB). All bacterial isolates were checked for their ability to produce indole-3-acetic acid (IAA). Out of 30, six Hg-resistant and NFB isolates, UM-3, UM-5, UM-7, UM-11, UM-26 and UM-28 were selected for high production of IAA as 18.6, 16.7, 16, 18.7, 14 and 16 µg/ml respectively (p<0.05). The biochemical and plant growth promoting characterization of selected Hgresistant and IAA producing NFB isolates was performed by using different tests such as Gram staining, morphological characterization, triple sugar iron (TSI), catalase, oxidase, HCN and H2S production and then phosphate and potassium solubilization. The 16S rDNA ribotyping and phylogenetic analysis of six highly Hgresistant and IAA producing NFB isolates was performed by PCR-amplification of 16S rRNA gene and characterized as Exiguobacterium sp. (KJ736011), Bacillus cereus (KJ736012), B. subtilis (KJ736013), Enterobacter cloacae (KJ736014) and Pseudomonas aeruginosa (KJ736016 and KJ736017). B. cereus UM-5 gave promising resistance against HgCl<sub>2</sub> (30 µgml<sup>-1</sup>) due to the presence of merB gene. The structural determination of MerB protein was carried out using bioinformatics tools i.e. Protparam, Pfam, InterProScan, STRING, Jpred4, PSIPRED, I-TASSER, COACH server and ERRAT. These tools predicted the structural based functional homology of MerB protein (organomercury lyase) in association with MerA (mercuric reductase) in bacterial Hg-detoxification system.

### ANTIBACTERIAL AND SYNERGISTIC EFFECTS OF MEDICINAL PLANTS AGAINST DRUG RESISTANT SALMONELLA PARATHYPHI A

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Salmonella enterica serovars para Typhi (S. Paratyphi A), belongs to family enterobacteriaceae is causative agent of enteric fever and contributes major role in mortality and morbidity. The current study was designed to study the phytochemical effect of selected medicinal plants and to determine MIC of Ceftriaxone and Ciprofloxacin alone against drug resistant S. paratyphi A as well as to assess the antimicrobial and synergistic effect of selected medicinal plant extracts in amalgam with the mentioned antibiotics. The resistant strains were obtained from Labortaey of Medical Microbiology, Department of microbiology, KUST and were screened for MIC determination of Ciprofloxacin and Ceftriaxone using Agar Dilution Method. Medicinal plants extracts along with antibiotics Ciprofloxacin and Ceftriaxone were screened for their synergistic effects against resistant isolates of S. paratyhi A. The data was analyzed by using two way ANOVA the P value <.05 was considered significant. It was determined from the current study that MIC of Ciprofloxacin ranges from 8 μg/ml to 256 μg/ml while Ceftriaxone ranges from 8 μg/ml to 512 μg/ml. From the antibiotic susceptibility analysis it was also concluded that no resistance was shown against levofloxacin. To check the antibacterial activity of plant extracts alone it was concluded that S. paratyphi A was very resistant to Descurainia sophia while no resistance was shown against Berberries spp.

# HERBAL MEDICINES AS POTENTIAL ANTI-INFLAMMATORY AND ANTIMICROBIAL SOURCES

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Frequent exposure of animals to antibiotics, to treat different ailments and as food additives for their rapid growth, is not only the source of multi-drug resistance in animals but it has also caused the exchange of resistant genes between animals and human beings. Moreover, persistent infections are also cause of inflammations that lead to the use of non-steroidal anti-inflammatory drugs (NSAIDs) as primary treatment that ultimately cause variou side effects e.g. ulcer etc. Being economic, safer and because of their hailing properties, natural plants and herbs have become the need of the day. Current study has been conducted to examine in vitro antimicrobial and antiinflammatory potential of ethanolic extracts of Mentha arvensis, Cymbopogon citratus and Camellia sinensis against E.coli, S. epidermidis, and S. aureus, isolated from wound, pus and infectious blood samples of infected animals and human. Isolates were identified by biochemical tests following standard manual. Identified isolates were tested against multiple antibiotics to check their susceptibility following CLSI guidelines and multi-drug resistant isolates were selected for further tests. Mentha Arvensis showed antimicrobial activity in an order S. aurous > S. epidermitis > E.coli. Camellia sinensis showed activity in an order S. epidermitis > E.coli > S. aurous. Cymbpogon showed mximum and equal activity against both S. epidermitis and S. aurous and least against E. coli. In-vitro antiinflammatory activity of sample extracts were used to prevent heat induced membrane lysis in erythrocytes and significant anti-inflammatory activity was shown by Mentha Arvensis followed by Cymbopogon citrate and Camellia sinensis. Aspirin was used as standard anti-inflammatory drug. All the experiments were performed in triplicates. Based upon the supporting results with the bioactive components of these plans, use of herbal medicines as antibiotics, anti-inflammatory drugs and feed additives is not only recommended for human beings but also for animals to avoid maximum exchange of multi-drug resistant microbes but also to avoid other side effects because of synthetic medicines.

### 5. MOLECULAR BIOLOGY

#### COMPARATIVE GENETIC DIVERSITY IN DOMESTIC STOCKS OF CATLA CATLA USING MICROSATELLITE DNA MARKERS

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The patterns of genetic diversity and genetic structure were analyzed by screening a total of fifteen microsatellite loci in five hatchery populations of  $Catla\ catla$ . Total 250 individuals (50 individuals from each site) of  $C.\ catla$  were collected and dorsal muscle tissues of the sampled fish were used for the genomic DNA isolation. In all the hatchery stocks of  $C.\ catla$ , low to moderate level of genetic diversity were found in terms of an average number of alleles and observed heterozygosity. The number of alleles average values were noted ranged from 2.800 to 4.000. In the examined populations, the values of effective alleles were measured less compared to the number of alleles. Expected heterozygosity values were recorded higher in comparison to observed heterozygosity. On the average base,  $F_{IS}$  values were found positive in all the examined populations, although some loci showed negative values. No deviation from HWE was observed. Low genetic differentiation between captive populations was indicated by the pairwise estimates of  $F_{ST}$ . Most of the variation was found within the individuals in the populations by applying AMOVA. Analysis of genetic relatedness among all the populations was estimated by constructing PCA, structure and neighborhood joining tree that showed major cluster patterns among the captive populations. Those populations that share the more genetic identity lies in the same cluster, while, populations with a lowest genetic identity fall in a different cluster. The present study inferences would be helpful in resolving the genetic problems relating to  $C.\ catla$  re-stocking plans and brood stock management practices.

### ASSESSING DISCRIMINATION EFFICIENCY OF MITOCHONDRIAL GENE COI FOR SPECIES/BREED LEVEL IDENTIFICATION OF GENUS *CAPRA*

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Short and standardized region of the mitochondrial genome has been revealed to be an effective method for molecular identification of organisms. The molecular taxonomy generates a public barcode library for the identification of different species. Composing a reference library of DNA barcodes is the master key for identifying species with firmer and inexpensive sequencing. A recent work suggests that 400-700bp region of the cytochrome c oxidase subunit I (COI) gene in the mitochondrial DNA could serve as a DNA barcode for the identification of animal species. This study tested the efficacy of a COI barcode in discriminating goat breeds, which has not been extensively tested. We access this gap by examining the performance of the COI gene for six different breeds of goat and identified all breeds as a *Capra hircus* (domestic goat). Based on 528 COI positions the average intraspecific genetic distance was (0.0016%) lower than the average interspecific genetic distance (0.03%), suggesting the discrimination efficiency of COI gene. The phylogenetic analysis of COI sequence revealed that *Capra sibrica* (Siberian ibex) formed the evolutionary basis of genus Capra. More analysis is required to illustrate the high COI gene divergence between the goat breeds. Analysis of the DNA barcode is urged as a faster, efficient, cheaper and easily available method for recognizing, analyzing and documenting the local goat breeds of the country.

### 1-AMINOCYCLOPROPANE-1-CARBOXYLASE DEAMINASE (ACCD) OF ASPERGILLUS FUMIGATUS: MOLECULAR MODELING AND IN SILICO CHARACTERIZATION

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Horizontal gene transfer (HGT) from prokaryotes to eukaryotes enable them to acquire entirely novel functions and enhances adaptation to changing environments. For instance, a prokaryotic gene acdS has been transformed to fungi. This gene encodes pyridoxal 5'-phosphate (PLP) dependent enzyme, 1-aminocyclopropane-1-carboxylase deaminase (ACCD) that transforms plant's ethylene precursor ACC to α-ketobutyrate and ammonia to modulate stress resistance. This study focuses on molecular modeling and in silico characterization of ACCD of Aspergillus fumigatus var. RP-2014 (KEY75690, 397 aa) using various sequence and structure based predictors and quality assessment of the proposed models. The structure consists of ten  $\alpha$ -helices, ten  $\beta$ -sheets and four  $3_{10}$  helices. The helices and folds can be further divided into two structurally dissimilar and interconnected subdomains. Catalytic residue (K66) and eleven PLP binding residues are also located. Out of four Cysteine residues, C204 and C230 are predicted to form the disulfide bond. The predicted molecular weight is 42885.11 and theoretical pI is 6.61. Molar attenuation coefficient (ε) at 280 nm is 41370 M<sup>-1</sup> cm<sup>-1</sup>, assuming all C residues are reduced. The instability index (35.51) classifies it as stable while GRAVY value (-0.146) indicates hydrophilic nature. RC plot indicated that about 83 % of residues lie in the most favored region that infers good quality of the proposed three-dimensional structure. The predicted LG score and MaxSub score were 4.849 and 0.332, respectively, which indicate correct model. A phylogenetic tree shows evolutionary relation among 14 Aspergillus species taken Pseudomonas spp. as outgroup with only 33 % sequence identity.

# DIAGNOSIS OF *MYCOBACTERIUM BOVIS* THROUGH CONVENTIONAL AND MOLECULAR AND TECHNIQUES

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A cross sectional study was carried out in order to establish a rapid and stable method for diagnosis of Mycobacterium tuberculosis infection through conventional and Molecular techniques and decrease the side effects of late diagnosis of patients and health system. Decrease long process of laboratory identification of the infection rate of *Mycobacterium bovis* and establishing new diagnosis methods was one of the reasons for this study. To accomplish this objective, collected m ilk and blood specimens from 100 BTB patients with clinical doubt of pulmonary tuberculosis were studied with both molecular and traditional techniques, Polymerase Chain Reaction (PCR), Enzyme-linked immunosorbent assay (Elisa) (IgG and IgM), Acid-Fast Bacillus stain (AFB) and culture methods were compared. The sensitivity and specificity of all methods were determined by using the PCR results as the gold standard. The overall sensitivity, specificity, positive predictive value and negative predictive value of AFB were 47.37, 49.38, 18.0 and 80.0%. These values for culture method was 66.67, 52.94, 20.0 and 90.0% and for IgG antibody were 70.0, 55.0, 28.0 and 88.0%, IgM antibody were 76.47, 55.42, 26.0 and 92.0% and for PCR were 100.0, 60.97, 36.0 and 100.0% respectively. It was concluded that maximum sensitivity and specificity can be achieved by PCR method.

### ASSOCIATION OF EXON 31 OF LRRK2 WITH THE ONSET OF PARKINSON'S DISEASE IN PAKISTANI POPULATION

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Parkinson's disease (PD) refers to the indelible and progressive neuronal regression that presents itself in patients with a variety of motor and non-motor symptoms. The etiology of this disease remains somewhat elusive; however, several studies indicate that it involves manipulation of genes by environmental factors. Mutations in at least 17 different genes are linked to onset of familial PD. LRRK2 mutations are considered to be the most classic genetic cause of both Sporadic and Familial PD. A total of 20 mutations are identified in this gene so far, out of which 6 are considered extremely pathogenic. Therefore, the aim of present study was to determine the association of polymorphism on Exon 31 of LRRK2 gene with the onset of Parkinson's Disease in patients of Lahore, Pakistan. Sampling was carried out along with DNA extraction and after Sequencing, Allelic and Genetic analysis was performed by using online SHESIS software. Haplotype analysis and Linkage disequilibrium was also conducted. Results indicated that males were at higher risk to develop PD at an early age than females and head trauma was a significant risk factor in PD onset among Pakistani population. However, mutations in Exon 31 of LRRK2 were not found to be significantly associated with PD in Pakistani Population.

### CO1 GENE-BASED IDENTIFICATION OF SCHIZOTHORAX SPECIES OF RIVER SWAT

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Fishes are considered as the most abundant class of vertebrates, there are more than 30,000 species are present worldwide. Fishes show vast variety in their morphology, in the habitats they occupy and in their life. Unlike the other frequently documented vertebrates, fishes are a diverse group. The *Schizothorax* also called snow trout is a genus of the Cyprinidae family. There are fifteen genera and more than hundred species are present throughout the world. The materials used in the current study cast nets, dragon nets, hooks, automatic rods, gill nets and hand nets. From each fish muscle tissue was taken from the right side of the caudal region and preserved in a separate falcon tube containing 99.8% ethanol. Identification of these fishes was done through different taxonomic and systemic keys and further conform through PCR. All collected *Schizothorax* fish were initially identified by key. Morphologically identification of fish is not 100% accurate so we identified the *Schizothorax* species with CO1 gene specific primer amplification through PCR.

# CLONING, EXPRESSION AND EFFECTIVE SOLUBILIZATION OF RECOMBINANT PCAL-0672

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An open reading frame (Pcal\_0672, WP\_011849356) of 485 amino acids residues corresponding to putative glycoside hydrolase of family 57 was cloned from *Pyrobaculum calidifontis* and expressed in *Escherichia coli* under the control of T7 promoter. Pcal\_0672 showed upto 81% sequence identity with the uncharacterized members of family GH57. However, low sequence homology (19%) was observed with the characterized thermophilic members of this

family. The expression analysis of the soluble and insoluble fractions after lysing the host cells revealed that recombinant Pcal\_0672 was produced as an insoluble aggregate in *E.coli*. Despite being produced in insoluble aggregates the recombinant enzyme was highly active when analyzed by zymogram. These insoluble aggregates were highly thermostable and retained their activity after boiling, in the presence of 1% Triton X-100 or 5% SDS. Further, its gene product was successfully refolded by taking expression at 17 °C temperature. Significant enzymatic activity was attained with its soluble fraction by using soluble potato starch. Pcal\_0672 is active member of class GH57 (having starch as its substrate).

# DETERMINATION OF THE SPECIES OF GENUS AUSTRUCA (FAMILY OCYPODIDAE) USING MITOCHONDRIAL (COI), RIBOSOMAL RNA (28S) GENES FROM THE PAKISTANI COASTAL AREAS

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Mitochondrial DNA (COI) and Nuclear DNA (28S rRNA) genes are genetic markers used as uniform and reliable evidence for the species level bio-cataloguing system compared with the morphological identification. Current study is based on the DNA barcoding and nuclear genetic analysis of genus *Austruca* from Sandspit, Pakistan. Earlier studies have recognised the intra-population morphological variability among the species of genus *Austruca*. The discrimination of species in the genus *Austruca* is particularly difficult due to some overlapping of morphological characters. During current study, each species of this genus was investigated through morphological and genetic data and compared with available sequences in Gene Bank. The conspecific and congeneric, K2P nucleotide deviation and nucleotide composition, number of haplotypes and haplotype diversity were estimated. The 5' region of cytochrome oxidase I (COI) and 28S rRNA gene were used to test the local sequences for identification of species along with gene bank sequences. Both sequences (Local and Gene Bank) found useful to discriminate and comparative for all the species tested. Genetic distance also showed higher level of variations between the two species (1.5% to 2.5%) using the 5' region showed higher level of intra-specific variation in isozyme (47% polymorphic loci) and molecular data (genetic distance = 1%). Further more studies are in process for further analysis from different localities and usage of different molecular markers (ITS-1 and ITS-2) to resolve this issue.

#### PROTEOMIC STUDY IN DEVELOPMENT OF QUALITY CONTROL OF SEAFOOD SAFETY

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Proteomics provides appropriate and powerful tools to investigate the key aspects of food quality and safety; it has made an important contribution to the discovery of biomarkers for food authentication. In this study, soluble protein and isozyme profiles with the help of commassie brilliant blue R-250 and Amylase were used as a marker for few selected doubtful species of shrimps; that were investigated as had dissimilar morphological identification as said by the seller and observed in the lab. Jaira; Indian white shrimps (name of all studied species added) were collected from Karachi and Korangi Fish Harbor. Pairwise comparisons of genotypes, based on the presence or absence of a unique and shared polymorphic product (bands), were used to regenerate Jaccard similarity coefficients and then used to construct the dendrogram by using the unweighted pair group method (U.P.G.M.A). A total of 19 bands was obtained through NATIVE PAGE analysis. SDS PAGE analysis showed 21 bands from 116.28 Da to 658.09 Da. Dendrogram showed *F. indicus* showed close genetic similarities in accordance with the morphological identification of *F. penicillatus*, while *F. merguiensis* showed in another cluster. The protein/isozyme fingerprint is useful to differentiate the shrimp species and can acts as a biochemical marker.

### GENETIC IDENTIFICATION OF SPECIES OF SEA SNAKES FOUND ALONG THE COASTLINE OF PAKISTAN

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As an important component of the marine habitats, sea snakes are understudied species. Although 14 species have been reported by various authors from Pakistan, but no further studies related to biology, venomics, molecular taxonomy and phylogeny of sea snakes have been conducted. Therefore, the current study has been designed to revive and authenticate the previous studies related to marine Snakes biodiversity using *Cyt b*, a mitochondrial gene. Five species, namely *Hydrophis caerulescens, H. schistosus, H. lapemoides, H. platurus* and *H. viperinus* were collected from three different localities, including Sonmiani (Baluchistan), Mubarak Village (Karachi) and Keti Bander (Sindh), were identified morphologically and genetically. Muscle samples were isolated from the tail region of all the species, which was followed by extraction and amplification of the gene. This was followed by Agarose Gel confirmation and sequencing. All the species were confirmed by GenBank with 99% similarity through BLAST search and procured data were submitted to GenBank.

# ESTIMATION OF PROTEIN CONCENTRATION IN TWO DIFFERENT SEASONS (PRE-MONSOON AND NORTH-EAST) IN MUSCLE OF TWO SPECIES OF SEA SNAKES (HYDROPHIIDAE) FOUND IN COASTAL WATERS OF PAKISTAN

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Sea snakes belong to group of creature called reptiles and are very unique and distinctive member of the marine environment. Almost 90 species of sea snakes are recorded all over the world, large number of Sea snake species are reported in the Indian Ocean. They are highly diverse in Northern Australia and East Asia as more than 25 species are reported in these areas. In Pakistani sea waters about 14 species has been reported. The current study based on biochemical analysis of protein in two different species of the sea snake (*Hydrophis caerulescens, H. schistose*) which collected from Sonmiani, Damb during two different seasons,i.e. Pre-Monsoon and North-East monsoon. Seasonal comparative study shows much variance in both species of sea snakes (*H. caerulescens, H. schistose*). The seasonal variability of protein concentration was found in both species and was highest (79.43±20.11) in *H. schistose* and (82.71±14.26) was found in *H. caerulescens* during Pre-Monsoon season and the low protein concentration was found in *H. schistose* (5.08±3.08) and *H. caerulescens* (4.27±2.05) during North-East season.

# DIFFERENTIAL SIGNALLING REGULATION OF TYPE I INTERFERONS THROUGH DISTINCT INTERFERON REGULATORY FACTORS IN RESPONSE TO VIRAL INFECTION IN FISH

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The interferons (IFNs) are a group of multifunctional cytokines which play important roles in immune system of vertebrates. Type I IFNs are considered as the major components of the innate immune response however their

ability to cope with different pathogens at the same time raise several questions regarding their differential regulation. The present study aimed to determine the differential regulatory mechanisms of type I IFNs in mandarin fish, *Siniperca chuatsi*. Initial gene expression analysis during poly (I:C) stimulation indicates that IFNh shows a rapid and robust induction in gene expression at 3 h (200 fold) and later by decreased in timely manner. In contrast, IFNc showed a delayed induction with peak at 12 h (115 fold) which indicates that both of these IFNs might be regulated through different components of the signaling pathway. This leads us to the search for the underlying mechanisms. *In-silico* analysis of the promoter regions shows the presence of two interferon regulator factor 7 (IRF7) binding sites in IFNc promoter region and two IRF3 and one IRF7 binding sites in IFNh promoter region. The expression analysis indicates that overexpression of IRF7 in MFF-1 cell can induce IFNc expression but not IRF3, however, IRF7 is not able to induce IFNh gene expression but the same shows upregulation during IRF3 overexpression in cells. Further the luciferase assay analysis shows the same pattern with IFNc and IFNh being regulated by two different IRFs, IRF7 and IRF3, respectively. This indicates that mandarin fish IRF3 and IRF7 can regulate differentially the expression of type I IFNs which is the basis for long term IFN activity to combat viral infections. Taken together, data indicates that the fish possess a complex antiviral mechanism to combat infection through regulation of distinct IFNs in differential timely manner.

# MUTATIONS IN POLYMERASE GENE OF HEPATITIS B VIRUS ASSOCIATED WITH HEPATOCELLULAR CARCINOMA AND LIVER CIRRHOSIS IN CHRONIC HEPATITIS B PATIENTS OF KHYBER PAKHTUNKHWA, PAKISTAN

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Hepatitis B is one the most common infectious diseases which can lead to Hepatocellular Carcinoma (HCC) and Liver Cirrhosis (LC) in chronic hepatitis B patients. Hepatocellular carcinoma (HCC) is a common tumour with worldwide distribution and HBsAg positivity in Pakistan is nearly 60% in cases of hepatocellular carcinoma. In Pakistan prevalence of HCC varies from 3.7%-16% of malignant tumors and most common cause of HCC is viral hepatitis B, C and D related cirrhosis. Similarly, approximately 87% of HCC is caused by viral hepatitis either C (68%) or B related cirrhosis (22%) This study aims to investigate the sequence of HBV P gene and the presence of P gene mutations associated with hepatocellular carcinoma among the study population. Blood was taken from 100 hepatitis B virus positive patients. DNA was extracted and HBV polymerase gene was amplified by PCR. Amplicon of 843 bp was sequenced and aligned with HBV genome sequences using BLAST. All the sequences were analyzed for mutations responsible for hepatocellular carcinoma and liver Cirrhosis. All the patients' genotype was D. Mutations related to HCC were detected in 40% of the study population. The mutations were found at positions L80G (3%), Y135S (2%), I169T (2.5%), V173L (2%), L180M (15%), A181T/V (15%), T184A/I/S (13%), S202I (17%), M204V/I (20%), I233V (12%), N236T (1%), M250V (1.5%) and I269L (1%). The mutations were prevalent in higher age groups. Elevated viral load was detected in most of the CHB patients. In conclusion, P gene mutations were found to be circulating in the study population. The frequently detected mutations are associated with a higher risk of HBV-related HCC in chronic hepatitis B patients.

### SEX DETERMINATION IN BIRDS: A MOLECULAR APPROACH

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Pakistan pet industry is growing with the advances in public socio-economic status. Breeding of commercial and domestic birds has emerged rapidly since last two decades. Several birds are monomorphic and become sexually dimorphic only in their adult ages. So, a difficulty has always existed to differentiate the sexes of birds during juvenile stages at the bird's sale markets and also during population and conservation studies. DNA base existing relies on chromodomain helicase DNA binding gene also referred as CHD is present on W chromosome and its

homologous Z chromosome. A PCR based sex determination method is optimized for distinct differentiation between male and female birds. A male bird sample produce a single 500 base pair amplification product whereas a female sample ends with 400 and 500 bps fragments after passing through amplification process. Conditions for amplification were optimized using DNA extracted from diverse samples of birds like blood, feather's base, cloacal and/or tracheal swab. After this comparative study we have found that tracheal and cloacal swabs are possibly the best option for sex determination of birds, as they pose very less harm to the birds as compared to the blood and feathers samples. It is suggested that the workers in open bird markets of Pakistan workers should practice collection of the swabs samples for birds gender determinations.

# PREVALENCE AND COMPARATIVE HAEMATOLOGICAL ANALYSIS OF *COLUMBA LIVIA* INFECTED WITH ECTOPARASITES IN DISTRICT KOHAT, KHYBER PAKHTUNKHWA, PAKISTAN

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Ectoparasites come under the classification of arthropods, may be obligatory parasites, programmed to feed on a living hosts to complete their life cycles. Several species of ectoparasites e.g. flies, lice, mites, ticks can infest C. livia. Ectoparasites affect bird health directly by causing irritation, discomfort, tissue damage, blood loss, toxicosis, allergies, anemia, elevated mortality, susceptibility to other infections, play a role as intermediate host, dermatitis and initiate excessive preening which in turn reduce the quality and quantity of meat and egg production, causes some of birds diseases such as Pasteurellosis, Newcastle disease etc. The present study was carried out from December 2018 to November 2019 with the objective of determining the prevalence of ectoparasites and to carry out comparative haematological analysis of non-infected and infected C. livia with ectoparasites in in district Kohat of province Khyber Pakhtunkhwa. The ectoparasites infesting C. livia were randomly collected by investigating different parts of their body and were preserved in 70% alcohol. Identification of ectoparasites were done using standard available keys under Dissecting microscope. For comparative heamotological analysis, 2 ml blood samples were collected from both infected and non-infected C. livia by venipucture of brachial vein and will be transferred to sterile glass bottles containing Ethylenediamine tetra acetic acid (EDTA) as anticoagulant. The data were analyzed statistically by SPSS, 2016 for chi square test. The overall prevalence of ectoparasites infestation during the study period indicated that 67 pigeons were positive for ectoparasites out of 253 pigeons. The sex wise prevalence was found to be highest in females (28.67%) than males (23.64%). The prevalence of ectoparasites was found to be higher in Jungle khel (35.56%) area of Kohat followed by Doda (29.23%), KDA (26.09%), main city (21.82) and Togh Bala (19.05%). In month wise prevalence, May (46.88%) show the highest prevalence and October (6.67%) and March (6.67%) show lowest prevalence. In the current study it was found that the RBC was 3.74±0.3 WBC was 46.2±3.1, Hb was 13.16±0.7, MCV was  $70\pm7.5$  and PCV was  $50.6\pm2.0$  in healthy male pigeon followed by  $3.58\pm0.3$ ,  $47.8\pm3.2$ ,  $12.86\pm0.7$ , 175.6±7.3 and 49±2.0 in infectd male pigeon. While in female population it was slightly differ showing as RBC was 3.46±0.4, WBC was 44.6±2.5, Hb was 12.08±1.1, MCV was 157.8±8.2 and PCV was 46.4±2.5 in healthy female pigeon followed by 3.32±0.3, 46.2±2.6, 11.7±1.0, 162.2±8.8 and 44.4±1.7 in infected female pigeon. Results of this study might be useful for controlling measures against the C. livia infestation with ectoparasites and subsequently improving the health conditions and growth.

# POPULATION DIFFERENTIATION THROUGH PROTEIN ELECTROPHORESIS IN OESTRUS OVIS (DIPTERA: OESTRIDAE: OESTRINAE) OF KARACHI, SINDH, PAKISTAN

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Oestrus ovis (nasal worm) is a dipteran insect belongs to sub-family Oestrinae. It is a world-wide parasite of goats and sheep mostly lives in the horns, frontal sinuses and nasal cavities of goats, and consequently leads to myiasis. This infection causes acute clinical manifestations, i.e. breathing disorders mucopurulent discharge, anorexia and weakness. Ultimately the pathogenic effects lead to considerably less animal production and significant economic losses. The current study was conducted to determine inter and intra population genetic differentiation in O. ovis five study area's population of Karachi. The samples were collected during December 2017 to March 2019 from the heads of slaughtered goats. The genetic variations were observed through the Polyacrylamide Gel Electrophoresis (SDS PAGE and Native PAGE) of General protein. The genetic variations were analyzed by Pop Gene software and revealed that the overall percentage of polymorphic loci within five populations ranged from 27.27% to 90.91%. The overall expected heterozygosity in all populations resolved through SDS PAGE and Native PAGE was  $0.181 \pm 0.096$  and  $0.153 \pm 0.099$ , respectively. The observed number of alleles (1.909-1.454), effective number of alleles (1.371-1.142) and Shannon's Information index (0.360-0.174) among all study areas population was determined. The gene flow within populations were found 2.065 and 2.002 in SDS PAGE and Native PAGE, respectively. The genetic identity ranged from 0.956 to 0.992 and genetic distance ranged from 0.008 to 0.045 observed through SDS PAGE, whilethe genetic identity and genetic distance observed in Native PAGE ranged from 0.968 to 0.988 and 0.012 to 0.033, respectively. The highest genetic identity and low genetic distance indicated that all study areas population were genetically close to each other and the gene flow was found among the O. ovis populations i.e.no sympatric and parapatric population differentiation was found and the populations were not reproductively isolated.

### PCR-RLFP BASED IDENTIFICATION OF PHLEBOTOMINE SAND FLIES IN DISTRICT KOHAT, PAKISTAN

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Several sand fly species of subfamily Phlebotominae are the vectors for intracellular *Leishmania* parasites which cause Leishmaniasis. It is important to study the distribution of sand flies to control Leishmaniasis in endemic areas. The current study was a PCR-RFLP of 18S r RNA gene based identification of sand flies to analyze the distribution of *Phlebotomus* sand flies in five different endemic areas of District Kohat region of Pakistan. A total of 532 sand flies were trapped by using CDC light trapes from April to August 2019. Morphologically all the sand flies were differentiated into two species of genus *Phlebotomus*. The two species were *Phlebotomus papatasi* and *Phlebotomus sergenti*. *Phlebotomus papatasi* was the prevalent (75.93%) sand fly in the proposed study area. Four different restriction enzymes (*HaeIII*, *HinfI*, *PstI*, and *AseI*) were used for the PCR-RFLP of 18S r RNA gene which produced DNA fragments of different sizes. The present study confirmed the previous studies conducted that a single restriction enzyme could not separate species belonging to the same genus or belonging to the different genera, therefore various restriction enzymes were used. Finally we conclude that morphological identification of sand flies is indespensible tool for their classification supported by molecular techniques like PCR-RFLP and DNA sequencing. Novel strategies for disease control and treatment require a better understanding of vectors diversity; therefore the identifications of both sand fly and *Leishmania* species are of great importance for the disease eradication in the endemic areas.

### BUTYRATE - A POTENTIAL ANTI-CANCER AND ANTI-DIABETIC DRUG *VIA* DECREASED EXPRESSION OF MTOR AND INCREASED EXPRESSION OF AMPK

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Sodium butyrate is a short chain fatty acid produced naturally by gut microbiota. It is a known histone deacetylase inhibitor with differential role in regulating genes' expression. Sirt1/AMPK and Akt/mTOR are major key markers for metabolic pathways involved in energy homeostasis and cell proliferation, respectively. In this study, effect of butyrate on expression of these genes in murine skeletal muscle tissue was determined and compared with that of metformin. For these mice (15-18 weeks) were divided in three groups; experimental (treated with butyrate), positive control (treated with metformin) and negative control (treated with buffer) groups. The doses were given orally as well as intraperitonially. After treatment, mice were sacrificed and skeletal muscle tissues isolated were stored at -80 °C. RNA isolated from each tissue was subjected to reverse transcription. Relative gene expression was measured by real time PCR. Expression of mTOR was found to be decreased and AMPK increased in response to both drugs irrespective of rout of administration. Thus, butyrate *via* decreased expression of mTOR can be manifested as an anti-cancer drug through decreased cell growth and proliferation. Butyrate *via* increased expression of AMPK can also be used as anti-diabetic as well as anti-obesity drug like that of metformin due to role of AMPK in energy homeostasis. NaB can also be involved in maintaining muscle mass and muscle regeneration by manipulation of mTOR and AMPK pathways.

#### USE OF PROBIOTICS FOR DISEASE MANAGEMENT

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Almost a hundred years have passed since theories on the prolongation of life by modulation of the intestinal ecosystem are being used. The beneficial effects of probiotic foods on human health are increasingly recognized by health professionals. Recent scientific work on the properties and functionality of living micro-organisms especially members of the genera Lactobacillus and Bifidobacterium, mainly used in food have suggested that probiotics play an important role in immunological, digestive and respiratory functions, and that they could have a significant effect on the alleviation of infectious diseases in children and other high-risk groups. Assessment of probiotic microorganisms depend on selection of strain specifically for human use, classification and identification of individual strains, defining and measuring their health benefits and their disorders associated with prevention of diarrhea caused by certain pathogenic bacteria and viruses, Helicobacter pylori infection and complications, inflammatory diseases and bowel syndromes, constipation, mucosal immunity and allergy. Others include urogenital tract disorders, bacterial vaginosis, yeast vaginitis, cancer, and cardiovascular diseases (CVDs). Probiotics naturally occur in chicory root, onion, leek, garlic, yogurt, buttermilk and some types of cheeses, and a number of supplemental forms are also available to consumers. However, there is no international consensus on the methodology to assess the efficacy and the safety of these products, at present. The requirements for good clinical studies have become even more important. Carefully controlled studies on selected strains could result in the future development of probiotic bacteria targeted for specific diseases and their prevention.

### AN EXPLORATORY RESEARCH TO GAIN A BETTER UNDERSTANDING OF THE MOLECULAR PATHWAYS INVOLVED IN NEUROINFLAMMATION

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Neuroinflammation represents a condition in which there is an activation of the resident immune system of the central nervous system (CNS). This can be either short-term or long-term with the latter often resulting in cell death

and neurodegeneration. One of the common events of both neuroinflammation and neurodegeneration is an increased concentration of pro-inflammatory cytokines. Consequently, understanding the regulation of inflammatory molecules is critical to gain insights into the biological and pathological events associated with them. Thus, this study was aimed at screening important molecular components i.e., transcription factors, Myo9b/RhoA and Wallerian degeneration pathways via utilizing bioinformatics and qRT-PCR tools to identify some of the key genes which might be contributing towards this process. Subsequently, the results of the current study indicated while the expression patterns of transcription factors as well as Myo9b are in line with their roles in causing neuronal death, however, the picture of Wallerian degeneration pathway is still elusive. And so, by identifying the molecular targets involved in the process of neuroinflammation, this study holds significance not only in presenting the targets for treatments but also for the development of a better understanding towards the underlying pathophysiology.

### 6. PHYSIOLOGY

### EVALUATION OF NEONATAL EXPOSURE TO BISPHENOL A AND ITS ANALOGUES ON THE SEXUAL DEVELOPMENT OF MALE RATS

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Bisphenol A (BPA) is one of the highly produced chemicals of the world used for the production of commonly used materials like food packaging, dental sealants and baby feeding bottles. Due to toxicity of BPA general public abandoned its use and manufactures introduced BPA analogues. The present study aims to investigate the possible reproductive effects of bisphenol A and its analogues BPB, BPF and BPS on testicular development in rats exposed during the neonatal stage of life. BPA, BPB, BPF and BPS were subcutaneously injected with different concentrations from postnatal day (PND) 1 to PND 10 in male rats. We found that BPA and its analogues were able to alter the hormone concentrations in the rats exposed neonattly and morphometrical results of testicular history showed alteration in the different cells of testis and epididymis. The above results suggest that neonatal exposure to different concentrations of BPA and its analogues bring about alteration in the reproductive system of male rats by both altering the hormonal system as well as testicular internal cellular morphology which indicates prominently that these BPA analogues are not that safe as claimed by the BPA industry.

# A HISTOLOGICAL AND BIOCHEMICAL APPROACH TO EVALUATE THE DIETARY EFFECTS OF BROILER AND DOMESTIC CHICKEN MEAT ON PUBERTY ONSET AND REPRODUCTIVE POTENTIAL OF FEMALE SPRAGUE DAWLEY RATS

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Increased consumption of broiler meat has led to the development of fast growing strain of broilers, which is associated with increase in fat and decrease in protein energy content. Since high fat energy has positive correlation with precocious puberty and related endocrine disorders, present study was designed to access the effect of broiler and domestic chicken meat consumption on puberty onset, oxidative stress and folliculogenesis in rats. For this purpose, adult Sprague Dawley rats were divided into five breeding cages. Control group fed on standard rat chow, second and third groups (B20 and B40) were provided with 20% and 40% of broiler chicken. Fourth and fifth groups (D20 and D40) were given 20% and 40% of domestic chicken. Feed was provided during gestation and lactation period and to their female pups after post-weaning period until Postnatal day (PND) 90. At PND 90, female rats were dissected out at estrous stage. Significant weight gain was observed in B40 group (P<0.01), B20 and D40 groups (P<0.05). Early puberty onset was evident in B40 (P<0.01), B20 and D40 (P<0.05) groups as compared to control. Malondialdehyde and ROS were significantly high in B40 group (P<0.05). Estradiol level was high in B20 and B40 group (P<0.05) and testosterone level increased significantly in B40 (P<0.05). Graafian follicles and Corpus luteum reduced (P<0.01 and P<0.05, respectively), while atretic follicles increased (P<0.05) significantly in B40 group. It is concluded that daily broiler meat intake is associated with early onset of puberty, increased oxidative stress and impaired folliculogenesis.

### AMELIORATIVE EFFECT OF OLIVE OIL AND POMEGRANATE ON OXYTOCIN INDUCED HEPATIC ANOMALIES IN MICE

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Oxytocin a neuropeptide hormone synthesize in hypothalamus used during parturition, birth and milk ejection but their ridiculous illegitimate used for cattle may produce anomalies. Study was conducted on forty, 3-4 months old male mice (*Mus musculus*) (30±3g). Animals were randomly divided as: Control (n=10) group, Oxytocin group: received 0.1ml/12h for 16 days, followed by without treatment for next 5 days, OT-PM and OT-OL were (0.1 ml/12h) pomegranate extract and olive for last 5 days respectively. The animals were euthanized by cervical dislocation on day 21<sup>th</sup>and ventricular blood was used for biochemical study and liver was used HE-stain. The histological section of Oxytocin group indicates the destruction of hepatocytes along with the sign of fibrosis and wide sinusoidal spaces along with accumulation of nuclei as compared to control. There were sign of lipogenesis and debris fills central vein along with dehydration in Oxytocin but Olive oil groups have more land marks of ameliorative effect and nullifying effects. The regulation of sinusoidal spaces also supports the regeneration ability as compared to Pomegranate.

# FEED PREFERENCES AND THEIR EFFECT ON BLOOD ANALYSIS AND MEAT ANALYSIS PARAMETERS IN MALLARD (ANAS PLATYRHYNCHOS)

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As the human population is increasing demand for milk, eggs and meat is also increasing exponentially and it is assumed that there might be 60% increase by 2025. The human demand for meat, milk and egg production is also increasing day by day and the situation is producing lot of pressures on livestock industry. The poultry yield in the tropics is very much increased due to high dynamics of development. Ducks are more disease resistant and their growth rate is faster than many of the commercial poultry. Duck meat is considered as a good source of providing protein (animal protein) and alternative source of chicken protein. The species which are mainly used to produce duck meat are Mallard (*Anas platyrhynchos*). The objective of this study was to evaluate the effect of different diet treatments on Mallard by morphometric measurements. The study was conducted on fifty-one-day old ducklings that were fed five different diets in same concentration. Ducks were randomly distributed into five groups each comprising ten ducks as follows:1st group (T0), 2nd group (T1), 3rd group (T2), 4th group (T3) and 5th group (T4) were given commercial feed, Wheat, Millet, Corn and Mixture of wheat, millet and corn (1:1:1) respectively for three months. Results showed that T4 contain significantly high protein and lower fat as compared to other groups and also higher blood serum protein and higher hemoglobin level as compared to other groups.

# ASSESSMENT OF HEAVY METALS (CADMIUM, CHROMIUM, COPPER, LEAD AND ZINC) CONCENTRATION IN FARMED FISH *LABEO ROHITA*, *CATLA CATLA*, AND *CIRRHINUS MRIGALA*

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To measure the heavy metals concentration in farmed fish. Three carp species were sampled from three different local fish farms of Bhone, district Jhang. Sample of fish were randomly collected from sampling station.

Five heavy metals, Cd (cadmium), Cr (Chromium), Cu (Copper), Pb (Lead) and Zn (Zinc) were analyses in four organs liver, muscle, intestine and gills of three carp species. Zn was found significantly high in all five metals and gills show maximum concentration of metals because of their direct contact with water followed by liver, intestine and muscle. Highest concentration of Zn was found in the gills of *Catla catla* (44.70±4.13) and the lowest concentration was found in the muscle of *Labeo rohita* (7.07±0.55). All the metals were found above the permissible limits set by FAO. This study proven that the fish cultivated in the farms of Bhone, district Jhang are not safe for human use it create severe health problems for consumers use these farmed fish.

### COMPARATIVE ANALYSIS OF EFFICIENCY OF BIODIESEL AND NANO FUEL EXTRACTED FROM DIFFERENT FISHES OIL

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In present time energy crisis is rising due to over population and as result petroleum products are depleting expeditiously. This scenario causes so many environmental hazards. These drastic situations divert the attention of world towards the renewable resources of energy as an alternate way of petroleum diesel as well as to cope with these climatic conditions. Biodiesel is gaining attention due to its unique characteristics like renewable, degradable, cheap and environment friendly fuel. In present study three different types of fish oil (Rohu, Mori and Tilapia) was used with four combinations (1:1:1, 2:1:1, 1:2:1, 1:1:2). Transesterification process was used to make biodiesel from four different combinations. Later on nanoparticles (ZnO) were added to check the effect of nanoparticles on biodiesel efficiency. Present study illustrate that the ratio which contained the double amount of Rohu fish oil showed best results of selected parameters which are used to check the efficiency of biodiesel. This combination showed good flash point, cloud point and pour point values, high calorific value as well as low density and kinematic viscosity then other combinations of fish oil. Furthermore, the addition of nanoparticles was also proved to be good in enhancing the efficiency of engine.

# EFFECT OF DIFFERENT DIETARY PROTEINS ON GROWTH PERFORMANCE OF PEKIN DUCK (ANAS PLATYRHYNCHOS DOMESTICA) IN CAPTIVITY

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The study was conducted to evaluate the effect of different dietary protein concentrations on body weight and morphometric traits of Peckin duck (*Anas platyrhynchos domestica*). For this purpose thirty day-one old female ducks were selected. They were fed *ad libitum* along with different experimental diets. Experimental diets were consist of varying levels of protein concentrations. The three levels (16.5%, 21% and 22%) of protein concentrations were prepared. Ducks were randomly distributed into three groups and received protein treatment i.e. group A (16.5%), group B (21%), group C (22%), for three months. The body weights and measurements of body parts were taken on weekly basis. The body weight (2281.78±7.7g), bill length (7.2±0.01cm), bill width (59.67±0.14mm), head length (16.01±0.03cm), neck length (15.98±0.06cm), wing length (23.70±0.02cm), wing span (75.34±0.05cm) and foot length distance (10.44±0.03cm) were noted significantly (p<0.01) higher in group C as compared to group A and group B. This study concludes that increased dietary protein concentration make growth performance better in Peckin duck.

## STUDY ON MILK YIELD, MILK COMPOSITION AND WEIGHT GAIN IN HOLSTEIN FRIESIAN DAIRY COWS USING THREE DIVERSE CONCENTRATE RATIO

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The aim of the current study was to evaluate the effect of three different concentrates on milk yield, composition, and weight in Holstien Fresian dairy cows in spring 2019 for a period of 60 days on mid lactating HF cows. The experiment carried out at the Dairy Farm of the Agriculture University Peshawar Nine HF dairy cows of almost similar lactation stage, daily milk yield and of parity 3 were selected. The cows were divided randomly into three groups A, B, and C. Each group of cows was fed with different concentrate rations to evaluate its effect on daily milk yield, composition, body weight gain and cost of concentrates per liter milk yield. Cows in control group (A) were fed commercial dairy feed (Aan vanda) along with green barseem (Egyptian clover) as a basal diet. The cows in group B were supplemented with formulated cottonseed cake (CSC) along with basal diet while cows in group C were offered formulated whole cottonseed (WCS). The concentrate rations offered to different experimental cows groups were iso-caloric and iso-nitrogenious. Highly significant (P<0.05) results were observed in DMY, milk fat%, milk lactose%, milk solid not fat%, milk total solid% and in cost of concentrate rations per liter milk yield among the three treated cows groups. High DMY (13.18ltr) was recorded in group C fed with formulated WCS, followed by group B (11.55ltr) fed with formulated CSC while low (10.52ltr) was noted in control group offered commercial dairy feed (Aan vanda). The group C supplemented with formulated WCS yielded 20.1% more daily milk than control group dairy cows. Fat content were significantly different in all three treated groups of dairy cows. Fat content (4.75%) was recorded in cows of group C followed by group B (4.26%) and (3.84%) in control group. Milk lactose% was higher (4.61%) in control group followed by group B (4.58%) and group C (4.39%) respectively. Increase in SNF% recorded in the milk of group B (8.82%) followed by group C (8.80%) while low (8.03%) in control group. The difference in SNF content is not significant between Group B and C while both groups have significant difference than cows in control group. The data trend indicates that TS% was higher (13.6%) in group C followed by group B (13.1%) and (11.6%) control group. Higher (Rs.14.5) concentrates cost per liter milk yield was observed in control group as compared to group B (Rs. 13.9) and group C (Rs. 12.7). The result of milk protein content was non-significant (P>0.05) among all the three treated groups. However, more (3.09%) milk protein was recorded in control group followed by group B (2.92%) and (2.91%) in group C respectively. Non-significant (p>0.05) difference was observed in live body weight gain among the experimental cow groups in the trial period. However, the mean changes in live body weight observed were (3.71kg, 2.69kg, 3.66 kg) in group A, B and C respectively. It was concluded that feeding formulated WCS have a significant effect on DMY, milk fat%, SNF%, TS% and cost of concentrates per liter milk yield Holstein Friesian Dairy Cows.

## HEAT SHOCK PROTEIN-70 IS EXPRESSED IN HIGHER QUANTITIES UNDER THERMAL STRESS CONDITIONS

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The cellular response to stress is mediated by intracellular proteins, called Heat Shock Proteins (HSPs). Among various known stressors, heat is a major factor that induces the production of HSP70. Keeping in view the very hot conditions of Dera Ismail Khan (D. I. Khan) division where the temperature remains at 45-50°C

during the months of June to September, it was hypothesized that heat stress conditions do induce the overexpression of HSPs, especially HSP-70. Is was thus attempted to find out the possible role of HSP-70 in those human being having the age of 90 years or above, called longevity people, against heat stress conditions, in comparison with control people with age below 60 years. Whole blood samples of 45 longevity individuals and 20 samples of control people were collected in D. I. Khan during September 2018 to October 2019. The study was approved by Gomal University ethical review board and written consent of each individual was taken prior to collection of blood sample. For serum collection, blood samples were centrifuged at 1000xg for 15 minutes. Quantitative measurement of HSP70 protein was determined using sandwich ELISA technique. The maximum serum HSP70 level observed was 42ng/ml and minimum serum HSP70 value was 13ng/ml, with median value of 28ng/ml. In control group, maximum concentration observed was 38ng/ml while the minimum serum level was 18ng/ml with median value of 13ng/ml. In longevity males, serum HSP70 levels increased in individuals between 89 to 91 years of age, peaked between 92 to 97 years but comparatively lower having the age of above 98 years. On the other hand, serum concentration of HSP70 in longevity females were highest in those having 89 to 92 years, lower in 93 to 97 years and, like males, lowest having age above 98 years. The study hence, showed that longevity individuals had higher quantities of serum HSP70 compared to control group, and again, males contained higher concentration than longevity females, thus confirms that heat stress induces the expression of HSP70 in both normal and longevity individuals but in longevity individuals this expression is slightly greater than normal individuals. Whether this increased expression has any impact on longevity is still not clear that needs to be deciphered.

### EFFECT OF PHYTASE SUPPLEMENTED MORINGA BY-PRODUCTS BASED DIETS ON THE PERFORMANCE OF OREOCHROMIS NILOTICUS FINGERLINGS

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This study was performed to develop an eco-friendly and low cost feed for 'Nile tilapia' fingerlings using Moringa derived products supplemented with phytase (PHY) enzyme. *Moringa oleifera* seed meal and leaf meal were used as the major components to make seven test diets (TD). These Moringa based test diets were developed by using different PHY levels (0, 500, 650, 800, 950, 1100 and 1250 FTU per kg). Tilapia were fed at the rate of 4% of their live weight two times a day. After 70 days feeding trials, blood samples from each replicate were collected for the analysis of hemato-immunological parameters. Then these fish were sacrificed and dried for carcass analysis. Maximum retention of protein (18.26%) and fat (8.92) were found at 950 FTU per kg in comparison to other test diets. Study of hematological indices revealed that the maximal values of RBCs (2.92×10<sup>6</sup> mm<sup>-3</sup>), PLT (64) and Hb (8.19 g/100ml) were recorded at 950 FTU per kg level. Similarly, highest MCH, PCV, MCV and Ht. were also observed at 950 FTU per kg level. For the study of expression of growth genes, gel electrophoresis method was performed. These results indicated that PHY added diets at 950 FTU per kg showed maximum improvement in hemato-immunological parameters and body composition of tilapia with no side effects on fish health and non-differentiable results regarding growth gene expression.

# DEVELOPMENT OF ECONOMICAL AND ECOFRIENDLY FEED FOR COMMON CARP (CYPRINUS CARPIO) FINGERLINGS FED ON PHYTASE SUPPLEMENTED MORINGA BY-PRODUCT BASED DIET

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Present study work was performed to develop the economical and ecofriendly diet for fingerlings of *Cyprinus carpio* by the help of phytase addition in *Moringa oleifera* by- products based diet for maximum growth performance. Several anti-nutritional substances are found in plant derivatives for example phytate, that make the nutrients and minerals unavailable to fish, hence leading to poor growth performance of fish. Combination of Moringa seed meal and Moringa leaf meal was utilized as test ingredient. Seven test feeds were made, containing graded levels of phytase (0, 500, 650, 800, 950, 1100 and 1250 FTU per kg). In feeding trial of 70 days, fish fingerlings were given feed two times in a day at 4% of wet weight of their bodies and faeces were collected. All studied parameters got better at phytase containing diets. Growth performance parameters i.e. weight gain; 25g, specific growth rate; 1.67 and feed conversion ratio; 1.10 were improved to maximum at 950 FTU per kg. Digestibility of nutrients (crude protein; 73%, fat; 71% and gross energy; 67%) was highest at 950 FTU per Kg. Similarly, minerals absorption was also maximum (Ca; 70%, Zn; 66%, K; 74%, Mn; 66% and P; 71%) at 950 FTU per Kg. Lowest growth efficiency, nutrient digestibility and mineral absorption were observed in fingerlings fed at control diet (0 FTU per Kg). Results proved that 950 FTU per Kg is the best level of phytase to produce economical and ecofriendly feed for *C. carpio* fingerlings as it decreases the discharge of minerals and nutrients in water bodies.

### STUDIES ON THE EFFECT OF FAT-SOLUBLE VITAMIN E AND K ON THE GROWTH PERFORMANCE AND ANTIOXIDANT ACTIVITY OF *LABEO ROHITA*

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Fish culture plays an important role for the benefit of people. In Asia and world carp contribute 70% of the inland aquaculture production. The research was designed to determine effect of the fat soluble vitamin E and K on the growth performance and antioxidant activity of *Labeo rohita*. Five diets were prepared as T1 control diet containing 0 mg/Kg VE and K, T2 containing 200 mg/kg VE, T3 containing 500 mg/kg VE, T4 containing 200 mg/kg VK, T5 containing 500 mg/kg VK. After acclimatization of *L. rohita* juveniles for few days they were transfer to ponds of Fisheries Research Farms, University of Agriculture, Faisalabad and fed twice a day for a period of six months. For better growth physico-chemical parameters were maintained at optimum level. *Labeo rohita* attained maximum growth during whole study period except December due to abrupt change in environmental factor such as temperature. Fish show maximum weight, fork and total length fed with T3 (VE=500mg/kg) that was 11.26g, 2.53mm and 2.96mm respectively. Feed intake, Feed conversion ratio and specific growth rate also increases with increment of vitamin E concentration. Due to high concentration vitamin supplemented diet increase in catalase and peroxidase activity while decrease in SOD activity was observed. Catalase and peroxidase show highest activity in T3 that was 134.03 U/mL and 0.37U/mL while SOD has higher in T1 (control) as 18.01U/mL. Statistical analysis determines the significant (p<0.01) fluctuation on growth and antioxidant activity of fish in whole study period.

## ANALYSIS OF ESTROGEN LEVELS AMONG HYPOTHYROID FEMALES DURING REPRODUCTIVE AGE IN RAWALPINDI, PUNJAB, PAKISTAN

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Hypothyroidism is an endocrine disorder characterized by low levels of thyroid hormones and is associated with reproductive problems predominantly in women. Estradiol is a sex steroid hormone and is involved in the development of secondary sex characters. The present study aimed to analyze the levels of estradiol hormone among hypothyroid females during their reproductive age. Blood samples were taken from normal and hypothyroid female patients. Blood samples of sixty normal and hundred hypothyroid females were taken from Rawalpindi, Punjab, Pakistan. Information including age, height, weight and any previous medical record was obtained with patients' consent. Exclusion and inclusion criteria were defined. The samples of control and hypothyroid patients were analyzed through ELISA to check hormonal concentrations. The estradiol levels were found to be significantly (P<0.05) decreased in hypothyroid patients as compared to control samples. The body mass index and TSH levels were found to be significantly (P<0.05) higher in hypothyroid patients than that of control samples. Present study concludes that decreased estradiol levels may be helpful in suggesting possible ways to treat infertility during hypothyroidism. Estrogen replacement therapies may be used in retrieving the complications induced by hypothyroidism.

### THE IMPACT OF SELENIUM AND VITAMIN E ON PHYSIOLOGY AND HORMONAL PROFILE OF DAMANI GOAT

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Goat has a significant role all over the world in increasing the economy of thousands of poor people, who earn their money by nurturing them in different environmental conditions. Nurturing Goat is a traditional profession of marginal farmers and landless workers in different region of under-developed countries. Through animal husbandry, farmer can produce food, fiber and some other raw material at very low cost from land that usually cannot be used to produce crop. The present study was conducted to find the effect of vitamin E and selenium (Se) on physiological and hormonal status of Damani goat in Dera Ismail Khan, KP, Pakistan during June 2018 to September 2019 under high ambient temperature. Forty Damani healthy, non-pregnant goats having similar initial body weight were selected. The goats were further equally divided into control and treated groups. The goats in the treated groups were supplemented with Se (0.3 mg) and vitamin E (50 mg) per kg of diet for 4 weeks. It was observed that respiration and pulse rate decreased significantly (P\0.05) on day 28 compared to the first day of the study in the treated group. The concentration of follicle-stimulating hormone (FSH) and progesterone were not affected but TSH, T3 and T4 were significantly (P\0.01) higher in the treated goats compared to the control. In the same way, total glucose, protein and cholesterol contents were significantly higher

in treated group of Damani goat compared to control. Moreover, cortisol and heat-shock protein-70 (HSP-70) increased significantly ( $P \setminus 0.05$ ) in Balkhi goat compared to Damani. It was thus concluded that vitamin E, in combination with Se, at the present doses improved the physiological, hormonal status in Damani goat.

### ASSESSMENT OF TRACE MINERALS ON OXIDATIVE STRESS DURING TRANSITION PERIOD IN CROSSBRED DAIRY COWS

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Trace mineral play significant role to overcome the oxidative stress during physiological process. Here we attempted to investigate the impact of trace mineral that includes Zinc, copper magnesium and selenium on the oxidative stress during transition period in the crossbred cows in the subtropical environment of Peshawar. Experiments were carried on six crossbred cows having approximately identical body weight and conditions with similar stage of gestation. Standard protocol was used for the collection blood and mineral analysis. Five ml of blood sample from each animal was collected at experimental interval. The collected blood specimen were centrifuge at 3000 rpm for 10 mints and the serum were kept until further analysis. Standard lab procedure was applied for determination of Cu, Fe, Mg and Zn. Likewise oxidative stress was measured via MDA concentration in serum as per standard procedure. The current study demonstrated that serum MDA concentration was significantly (P<0.05) higher in transition period two prior to calving in the cross bred cows in comparison with at calving or two postcalving period. our results further demonstrated significant decrease in the concentration of trace mineral including Zinc, CU, Mg, SE from two earlier to parturition until calving followed by gradually increase trend in the contraction of theses trace mineral. Thus it could be deduced from the current study that trace minerals have substantial impact on oxidative stress (MDA) during transition period in Cross breed dairy cows in the subtropical environments.

# HEMATOLOGICAL APPRAISAL OF ANAPLASMOSIS AND USE OF DIVERSE CHEMOTHERAPETIC AGENTS IN SMALL RUMINANTS

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Anaplasmosis is worldwide distributed tick-borne disease of ruminants associated with great economic significance. The object of the present study was to identify Anaplasmosis using microscopic examination with Giemsa Stained thin blood smear procedure in the small ruminants in relation with sex and breed and to study the effect of anaplasmosis on the Hematological indices in conjunction with treatments of the disease using different therapeutics agents in Mandi Bahauddin District. Experiments were carried on four hundred small ruminants including two hundred sheep and two hundred goats in the region selected on general criteria for the susceptibility of the Anaplasmosis. Sheep used were belonging to three breed including Salt Rang breed (85); Thali breed (53); Kaji

breed (35) and mixed or non descript sheep breed (27). Likewise goats used during current study were belonging to two local breeds namely Teddy (102); Beetle goat breed (67) and mixed or nondescript goat breed (31). Standard protocol was used for collection of the blood and thin blood smear preparation and hematological indices. For restorative trails 30 positive small ruminants were chosen in three different groups with three different therapeutic agents namely Oxytetracycline, Imidocarb diapropionat, and Diaminazin aceturate. The result of the current study demonstrated 54.75% prevalence of anaplasmosis in the total small ruminants (219/400) studied. Comparative percentage prevalence of anaplasmosis in sheep and goats was recorded as 61.50% and 48.0%. Relative percentage prevalence was higher in the female of both sheep and goats when compared with their corresponding male. Breed wise prevalence in sheep was recorded as 61.82, 54.17 and 52.38% in Salt range, Mix breed and Thali. Likewise in goat, prevalence was 70.97%, 46.08% and 40.30% in Mix breed, Teddy and Beetle breed. Furthermore, the hematological finding of the current study demonstrated considerable decease in HB level, PCV and RBC level in infected animal when compared with normal. Regarding the restorative trails, finding of the current study demonstrated that Oxytetracycline was highly effective against Anaplasma ovis followed by Imidocarb diapropionat and Diaminazin aceturate in goats and sheep. Thus it could be deduced from the current study that Anaplasmosis via A. Ovis is subsist abundantly in small ruminants in and around Mandi Bah Ud Din district, Punjab, Pakistan, have marked impact on the hematological indices. Furthermore, it was also inferred from this study that Oxytetracycline was the best medication against Anaplasmosis in both sheep and goats in the study area.

### EFFECT OF NATURAL GROWTH PROMOTERS ON BROILER GROWTH PERFORMANCE

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Feed plays crucial role in expansion of poultry industry. Extensive use of Antibiotic Growth Promoters (AGPs) in broiler feed led to the development of resistant microbes. Due to potential threat to human health, alternative strategies are required. Natural Growth Promoters (NGPs) can be effective alternatives to AGPs. The aim of the current study is to evaluate the effect of NGPs on growth performance, carcass characteristics, serology, intestinal morphology and protein profile of broiler chicken. For this purpose, 210 chicks were divided into 5 groups and were fed with starter (day 0-21, CP 22.3%, ME 2920 kcal/kg) and finisher diet (day 22-42, CP 19%, ME 3050 Kcal/kg). Feed treatments were formulated: Control (Basal), AGP (0.5% Zinc bacitracin), Prebiotic (0.2% MOS), Probiotic (0.1%) and Synbiotic (0.2%Pre+0.1%Pro). Growth performance data was collected on weekly basis. On day 42 blood samples for biochemical analyses, intestine samples for histomorphology and tissue samples for protein profile were collected. Intestinal morphology was analysed by ImageJ software. Protein profile was analysed by SDS-PAGE whereas images were analysed by ImageLab software. The results showed significant improvement in final body weight, BWG and FCR by Synbiotic group (p<0.05). Evaluation of carcass characteristics showed significant increase in dressing % by Synbiotics followed by Probiotic and Prebiotics. Serological tests revealed that NGPs can significantly decrease cholesterol and LDL whereas on the other hand NGPs were able to increase HDL significantly. It was observed that NGP fed broilers had more healthy intestine as compared to AGP. The NGP fed groups showed significant increase in villi length, crypt depth and villi to crypt ratio. It was also observed that Control and NGP increased tunica muscularis thickness as compared to AGPs. On SDS-PAGE different banding patterns were obtained (10-250kDa). It is concluded that NGPs might be efficient alternative to AGP and are more effective when fed in combination (Synbiotic).

### SEASONAL AND SEX RELATED HEMATOLOGICAL VARIATION AND SERUM ANALYSIS OF LABEO ROHITA FROM KOTRI BARRAGE NEAR JAMSHORO

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The present investigation on Seasonal and Sex related hematological variation and serum analysis of Labeo rohita from Kotri Barrage near Jamshoro was initiated from March – May, 2016 and from November, 2016 to January, 2017. The various haematologial parameters were taken into account and concluded that Male of L.rohita was found with higher values of Erythrocyte count (Mean of summer – winter 2.09 - 2.26), Leucocytes Count (Mean of summer – winter 164.90 - 145.55) and hemoglobin concentration (Mean of summer – winter 10.34 - 7.06), during summer and winter of that of female. The female of L.rohita was found with higher values of Erythrocyte Sedimentation rate (Mean of Summer – winter 2.38 - 2.56). The rest of parameters did not show any significant variation between sex during summer and winter season.

#### PSEUDO-HYPERTENSION

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A large proportion of hypertension is easily manageable through Homeopathic system of treatment and after sometime no longer issue exists while in Allopathic system life-long antihypertensive medication may be required. I used the term PSUEDO-HYPERTENSION for this kind of high blood pressure. This type of hypertension is most prevalent in youngsters and middle aged peoples suffering from diabetics and mentally depressed ones. This type of hypertension can be diagnosed and differentiated from common type of hypertension while checking blood pressure through careful observation for kind and quality of the sound noticed through stethoscope. In usual routine of blood pressure observing the sound noticed by the physician displays a specific standard reverberating echoing pitch like drum beating sound with high volume, while the same sound of the persons of PSEUDO-HYPERTENSION observed through stethoscope is of poor quality pitch, echo and volume. These sort of weak sounds are produced owing to drainage and deficiency of Vital Nutrients from the body along with the cells of the circulatory system. These nutrityely poor cells of circulatory system are forced to fulfill the demands of the body's energy requirement, this deficiency is indemnified by exerting over work thus producing high blood pressure or hypertension. As this sort of blood pressure is easily manageable (through homeopathic medicines) and possesses no firm ground for hypertension I used a new term for it as PSEUDO-HYPERTENSION. These high blood pressures or Pseudohypertension are found in majority of youngster, diabetics and psychogenic depressed persons. In teen ager boys the cause is self-abuse by onanism or masturbation (The young boys of this bad issue not only suffer from this habit but also have an internal drive for the propagation of this problem leading to the inflammation of prostrate gland. This inflamed prostrate gland makes genitalia extremely sensitive to touching materials, even touching of the cloths are not tolerated and seepage or leakage of prostrate secretion starts.) and in girls this is due to leucorrhoea( The leakage of fluid from the Uterine wall is faced by the girls in leucorrhoea.). These adverse situations make the body nutritively hollow, which force circulatory system to exert extra force to meet the demands of the body, which results in hypertension. The other group of peoples are Diabetics whose cells are hungry because of storage of insufficient amount of glycogen in the cells. On the other hand mentally tense or depressed people lack required nervous driving force for accomplishing body demands. Each and every problem needs its specific means of treatment. While in homeopathy, some times, same problem in different persons need separate type of medicine. Few typical medicines are given for guide to help in solving the problems. Bufa rana 30 (suppresses bad masturbative drive, it supports heart) Selenium metallicum 30 (cures inflammation of prostate gland or prostatitis and exhaustion of old age) Pyrogenum 200 (suppresses infection)

Hepar sulf 200 (fights deep infection)

Sepia officinalis 200 (medicine of women not girls)

Ignatia amara 6x / 30 (medicine of emotional ailment)

Veratrum album 6x / 30 (the best medicine for strengthening nervous system)

Homeopathic medicines that help to strengthen cytophysiology of Diabetics are as follows; (these are in three combinations).

Argentum nitricum 30 + Uranium nitricum 30

Acid lactic 6x + Acid phos 6x

Ginsing Q + Ginkgo biloba Q (its use may reduce or increase blood pressure, if it increases blood

pressure, only it, 3<sup>rd</sup> combination, should be discontinued as it is extremely powerful group of medicines)

Crataegus oxyacantha Q (it is general heart tonic, it provides rhythmic force to heart muscles)

## SUPPLEMENTATION OF POLYVINYLPYROLIDONE IN EXTENDER IMPROVES POST THAW QUALITY OF CRYOPRESERVED BUFFALO SEMEN

Bushra Rafaquat<sup>1</sup>, Rabea Ejaz<sup>2</sup>, Saima Qadeer<sup>3,\*</sup>, Asma Ul Husna<sup>1</sup>, Muhammad Sajjad Ansari<sup>4</sup>, Bushra Allah Rakha<sup>5</sup> and Shamim Akhter<sup>1</sup>.

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The study was planned to evaluate the effect of Polyvinylpyrolidone (PVP) supplementation in extender on cryopreservation of buffalo semen. The semen samples were collected from three Nili-Ravi buffalo (*Bubalus bubalis*) bulls using artificial vagina at weekly intervals for a period of three weeks (replicates; n=18). After collection, semen was immediately transferred to laboratory for initial evaluation. Qualified semen ejaculates (>1mL volume, >60% motility, >0.5billion/mL concentration) were split into four aliquots and diluted in *tris*-citric acid extender containing 0 (control) 0.01, 0.05 and 0.1mM of PVP. Semen was stored at 4°C for 2 h, kept at 4°C for 4 h, filled in French straws at 4°C, plunged and stored into liquid nitrogen container at -196°C. Semen straws were thawed at 37°C for 30 sec. and assessed for sperm motility, plasma membrane integrity, viability and chromatin integrity. Sperm motility and plasma membrane integrity were significantly improved in extenders containing 0.1mM and 0.05 mM of PVP compared to other treatment and control (P< 0.05). Sperm viability was equally improved in all experimental extenders containing PVP compared to control (P< 0.05). In conclusion, PVP supplementation at 0.1mM increased the post thaw quality of Nili Ravi buffalo semen.

## EFFECT OF IN OVO HONEY SUPPLEMENTATION ON HATCHIBILITY AND POST-HATCH PERFORMANCE OF BROILERS

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Research have shown that supplementation of exogenous nutrients to growing chicken embryo have significant positive effects on fertility parameters and post-hatch performance of chickens. The present study aimed to explore the effect of *in ovo* supplementation of honey on hatchability and post hatch performance of broiler chickens. Chicken embryos were inoculated with 500µl of 20% diluted honey while control group was inoculated with same

quantity of normal saline on day-15 of incubation. Upon hatching, day-old chicks of both honey treated and control group were shifted to experimental farm to explore their post hatch growth performance. The results exhibited that in ovo honey treatment significantly (P < 0.05) improved the hatchability percentage as well as birth weight of chicks as compared to control group. Furthermore, honey administration improved (P < 0.05) the feed intake, body weight gain and carcass weight while, reduced (P < 0.05) the feed conversion ratio and mortality in broiler chickens as compared to control group. These results established that in ovo supplementation of honey had better effect on hatchability and post hatch performance of broiler chickens.

# EVALUATION OF BLOOD BIOCHEMICALS, MORPHO-FUNCTIONAL EVALUATION AND CELL CYCLE GENE EXPRESSION IN KIDNEY OF GOAT FED SELENIUM-DEFICIENT DIET

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The present study assessed the effect of selenium deficiency in diet on histomorphology and antioxidant status in kidney of goat. A total of ten goats were randomly assigned into two groups and fed either Se-deficient diet (A, n=5) or Se-supplemented diet (B, n=5). The Se level in diet of animals in group A was 0.03 mg Se kg<sup>-1</sup> diet, whereas the animals in group B received same diet with supplementation of organic Se (selenium yeast) at the rate of 0.3 mg Se kg<sup>-1</sup> diet. Animals were fed experimental diets for a period of 10 weeks. At the end of experiment, the blood samples were collected to determine the biochemicals. After slaughtering, kidneys were extracted and two tissue samples were collected to determine the oxidative stress parameters and cell cycle gene expression, respectively. The results revealed that serum glucose and total protein levels decreased significantly (P < 0.05) in goats fed Se-deficient diet (A) compared with those fed Se-supplemented diet (B), however, urea and creatinine levels were not different (P > 0.05) between the two groups. The malodialdehyde (MDA) level significantly increased (P < 0.05) and the activities (U/ml) of glutathione peroxidae (GSH-Px) significantly decreased (P < 0.05) in A compared with B. The mRNA expression of cyclin D1 and CDK4 significantly decreased (P < 0.05) in A compared with B. The results of present study demonstrated that the selenium deficiency in diet altered blood biochemicals, induced oxidative stress and reduced proliferative gene expression in kidney of goat.

### IN VITRO SUPPLEMENTATION OF FATTY ACIDS IN EXTENDER FOR CRYOPRESERVATION OF BUFFALO SEMEN

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Present study was designed to evaluate the effect of supplementation of combination of fatty acids in extender for cryopreservation of buffalo semen. For this purpose, semen was collected from three adult buffalo bulls (Bubalus bubalis) of similar age by the help of artificial vagina ( $42^{\circ}$ C) for a period of 5 weeks (replicates; n=30). After collection, the qualified semen ejaculates (>1 mL volume, >0.5 billion per mL conc., >60% motility) were diluted in tris citric acid extender at 37°C. Six different experimental extenders were prepared containing fatty acids (arachidic acid 20 ng/mL,  $\alpha$ -linolenic acid 5.0 ng/mL and cholesterol 5.0 ng/mL) alone and in combinations. The extender without fatty acid supplementation was considered as control. Diluted semen was cooled to 4°C in 2 hours and equilibrated for 4 hours at 4°C. Cooled semen was filled in 0.5 mL straws at 4°C, kept on liquid nitrogen vapours for

10 min. and plunged in liquid nitrogen for storage. Thawing of frozen semen was performed after 24 hours at 37°C for 30 seconds and assessed for semen quality. Supplementation of combination of fatty acids in extender didn't improved sperm motility, plasma membrane integrity, viability and chromatin integrity of cryopreserved buffalo semen compared to supplementation of fatty acids alone in extender. In conclusion, addition of fatty acids in combination to semen extender was not found beneficial to improve quality of cryopreserved buffalo semen.

## FORTIFICATION OF SALT COUPLED WITH IODINE AND IRON: TO REHABILITATE INADEQUACY OF IRON AND IODINE

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Salt Fortification coupled with iodine and iron complex has been scrutinized. In many populations worldwide, Iodine deficiency disorder (IDD) are still a major public health concern Iodine-iron inadequacy subsist in emergent nations can be rehabilitate by fortified salt coupled with iodine and iron. Efficacy of iron fortified salt has been scrutinized among day-pupils and in divergent communities. The inclusion of iodine to iron fortified salt was thoroughly unstable as stated by primary evaluation. As a consequence, a stabilizer sodium hexametaphosphate has been used to generate new formula. It stabilizes fortified iron and iodine very well at a level of 1%. Coexistence of anemia and goiter could be cured by using salt coupled with iodine and iron. For the global establishment and to prevent the domino effects, rehabilitation of iodine inadequacy is of tremendous significance. Causes of brain damage precisely contain the inadequacy of iodine. The intellectual aptitude of day-pupils, life in divergent communities and economic fertility effected due to this mental inadequacy. Still multitudinous appraisals have been scrutinizing to surpass the Permanence and utilization rate of fortified salt coupled with iodine and iron.

## ASSESSMENT OF BULL SEMEN FERTILITY BY IN VITRO FERTILIZATION TESTS IN NILI RAVI BUFFALO BULL

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Bulls differ in their ability to produce offspring and bull variation is considered an important factor influencing *in vivo* and *in vitro* fertility rates. Therefore, precise selection of sperm from number of bulls is vital in order to categorize appropriate semen. To test male fertility by field trials is expensive and time-consuming. The present study aimed at determining outcome of different *in vitro* semen evaluation tests in order to achieve a better correlation between test results and fertility of bulls. For this purpose variation in progressive motility, plasma membrane and acrosome integrity, viability, sperm morphology, DNA integrity of cryopreserved semen and timing of the first cleavage was studied from six Nili Ravi buffalo bulls. Semen was collected in artificial vagina. The qualified semen ejaculates (>1 mL volume, >60% motility, >0.5 billion/mL concentration) was diluted in Tris citric acid extender, cryopreserved and assessed for semen quality. *In vitro* fertilization was performed following maturation of COCs collected from abattoir. Approximately 20 h after insemination with semen from one of the six bulls, presumptive zygotes was transferred to *in vitro* culture media and number of cleaved oocytes was recorded for each of the six bulls at 24, 28, 32, 36 h. Data of all semen quality assessment tests shows that sperm progressive motility

did not vary (P>0.05) among six buffalo bulls. Results of plasma membrane and acrosome integrity, viability, sperm morphology, DNA integrity and timing of the first cleavage showed that Bull 4 was found to show significantly better results. Field fertility also showed that bull 4 has higher field fertility. Semen quality assessment tests and timing of the first cleavage post insemination can be used to discriminate between bulls of high and low bull fertility. The study will assist to enhance the fertility rate of cryopreserved Nili Ravi buffalo bull semen not only during *in vitro* fertilization (IVF) experiments but also in *in vivo* trials.

# ASSESSMENT OF CHANGES IN SERUM CORTISOL LEVEL AND ESTIMATION OF LEAD IN TRAFFIC POLICE TOLERATING URBAN POLLUTANT STRESS LAHORE, PAKISTAN

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A longtime exposure to work place stress and urban pollutants in traffic police can lead to an elevated serum cortisol and lead level, which in return can lead to high blood pressure. In this study, the level of serum cortisol (ng/mL) and lead was assessed. A total of 64 samples were collected from subject (traffic police) and control groups in the morning as well as in the evening. The serum cortisol level determined by ELISA for all the subject groups including smoker and non-smoker groups was significantly (p<0.05) higher as compared to the control. The mean serum cortisol level in the smoker group was found significantly (p<0.05) higher as compared to the non-smoker group. The mean diastolic/systolic blood pressure (mmHg) of the subject, smoker, non-smoker and the control groups observed in the morning was 84.80/137.30, 86.60/142.62, 82.80/137.80 and 78.60/126.30 respectively. Similarly, in the evening it was 79.80/131.60, 82.52/137.80, 77.90/133.50 and 74.50/118.30 respectively for all the above groups. The level of lead estimation by atomic absorption spectrometer was found to be higher in all subject groups as compared to control groups. The blood lead level of subject group was significantly higher as compared to the control. Increase in the blood lead level was noted with increase in the time duration period. There was close correlation between blood lead level and blood pressure. Work place stress, in particular environment, to which traffic police is basically exposed, was found to be the main factor for the observed results.

## EFFECT OF SELENIUM ON OXIDATIVE STRESS AND PROLIFERATION DURING RAIN ADAPTATION IN COLON OF GOAT

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The present study assessed the effect of selenium on oxidative stress and proliferation in colon of goat fed high grain (HG) diet. Eighteen cross-bred goats, allocated into three groups (n=6/group) were fed low grain (LG, roughage: grain ratio 65:35), high grain (HG, 35:65) or HG diet supplemented with Se (HGSe) diets. The Se was added at the rate of 0.5 mg.kg<sup>-1</sup> diet from selenium yeast. At the end experiment (10 weeks), the animals were slaughtered and samples were collected. The volatile fatty acid (VFA) concentrations in colon fluid were determined by gas chromatography. Histo-morphology of colon tissues was carried out on HE stained slides. Antioxidant enzymes and MDA level in tissue was assessed through commercially available ELISA kits. The expression of proliferative genes was measured through qPCR. Results revealed proportional rise in total VFA concentrations and decrease in pH (P < 0.05) in HG and HGSe compared with LG. Histomorphological evaluation showed damages in the colon epithelium of HG, whereas HGSe attenuated such results. The malodialdehyde (MDA) level significantly increased (P < 0.05) and the activities (U/ml) of glutathione peroxidase (GSH-Px) significantly decreased (P < 0.05)

in HG; however, Se supplemented HG diet improved antioxidant status by alleviating such changes. The mRNA expression of cyclin D1 and CDK4 significantly decreased (P < 0.05) in HG compared with LG, whereas HGSe attenuated the expression of these genes and showed proliferative effect in colon epithelium. The results revealed that Se supplementation under stress condition reduced histomorological damages, improved antioxidant status and enhanced proliferative effects in colon of goat.

### EFFECT OF SELENIUM AND VITAMIN E ON GROWTH AND IMMUNITY OF RAINBOW TROUT (ONCHORHYNCHUS MYKISS)

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Feed having different concentrations of selenium and vitamin E was given to fingerling of Rainbow trout (Onchorhynchus mykiss) for 90 days at Trout Culture and Training Centre, Madyan Swat. The experimental feed contained two concentration (3mg/kg, 6mg/kg) of selenium in T1, T2 had two concentrations (160mg/kg, 280mg/kg) of vitamin E, while T3 had mixture of both selenium (1.5mg/kg, 4.5mg/kg) and vitamin E (80mg/kg, 90mg/kg). All these treatments were accompanied by group fed with controlled diet. Each concentration group had three replicates. The water quality parameter were checked on daily basis. The final weight of fishes was measured at the end of experiment for growth studies. Blood sample was taken from all the groups for hematological analysis and enzyme activity tests. A challenge trials was applied at the end of feeding trials with Aeromonas salmonicid bacteria. The data was subjected to analysis of variance (ANOVA) using SAS 9.0. The results showed that fishes from treatment 3 had the highest final weight, NWG, SGR significantly (p<0.05) greater than control group and other treatment groups as well while lowest FCR significantly (p<0.05) less than other treatment groups. Fishes fed with Se (3mg/kg), 6mg/kg) showed the lowest (p<0.05) NWG and SGR while FCR ratio was highest. The hematological parameters like RBC, Hb, MCV, MCH, MCHC, HEMATOCRIT and WBC also showed similar trends and was significantly (p<0.05) higher in fishes fed with mixture of selenium and vitamin E whereas fishes fed only with selenium had significantly low amount of these indicators. The bacterial challenge trials showed the opposite results. The mortality was highest in control group followed by fishes fed only with selenium while fishes fed with both selenium and vitamin E had significantly (p<0.05) lower mortality than control and other group.

# PREVALENCE OF RHIPICIPHALUS MICROPLUS INFESTATION IN LARGE RUMINANTS OF DISTRICT LAKKI MARWAT, KHYBER PAKHTUNKHWA, PAKISTAN

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Rhipicephalus microplus (formerly Boophilus microplus) is most important tick parasite of livestock throughout the globe. It is a hard type of tick belongs to the family Ixodidae and can be found on several hosts i.e., buffalo, cattle, donkeys, horses, goats, deer, dogs, pigs sheep and other wild animals. The present study was carried out from December 2018 to April 2019 with the objective of determining the prevalence of Rhipicephalus microplus tick infestation in livestock of district Lakki Marwat, KP Pakistan. The sample was collected by handling from different livestock in district Lakki Marwat and preserve in alcohol. Ticks were identified on the basis of morphological features. The data were analyzed statistically by SPSS, 2016 for chi square test. The overall prevalence of Rhipicephalus microplus tick infestation during the study period indicated that at overall 128(19.51%) were positive for Rhipicephalus microplus tick out of 656 samples. The sex wise prevalence was found to be highest in highest in females (29.6%) than males (14.33%). The Host wise prevalence of Rhipicephalus microplus tick infestation highest in buffaloes (69.69%), followed by cow's (49.76%). The prevalence of Hyalomma ticks was found to higher

(46.05%) in the age of above than 5 years followed in order by 1-5 years age (42.11%) and 1 year (27.8%) livestock. In different areas of Lakki Marwat different infestation rate was found in which Landiwa show the highest prevalence (63.97%) as compared to Ghazni Khel (28.94%), Titter Khel (27.05%), Aba Khel (15%) and Kaka Khel (13%). It is suggested that highly modified strategy are required to control the *Rhipicephalus microplus* tick infestation in livestock to increase the livestock yields and reduce the economic loses.

### COMPARATIVE STUDY ON LEVELS OF LIPID PROFILE IN THYROID PATIENTS AND NORMAL PERSONS OF KHAIRPUR CITY SINDH

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The thyroid gland is main endocrine organ in humans. The follicular cells of the thyroid gland secrete triiodothyronine (T3) and thyroxine (T4) hormones to control metabolic activities of body. Thyroid hormones have significant effects on synthesis, mobilization and metabolism of lipids and they are also involved in the regulation of lipid and lipoprotein metabolism. The altered secretions of thyroid hormones generate abnormal condition known as hypothyroidism and hyperthyroidism. Hypothyroidism is underproduction of the thyroid hormones tri-iodothyronine (T<sub>3</sub>) and thyroxine (T<sub>4</sub>) and excess production of T3 and T4 is called hyperthyroidism. Both abnormal conditions can have adverse effects on synthesis, mobilization and metabolism of lipids. In this response current research work was designed to compare lipid profile of normal persons and thyroid patients of Khairpur city. The data of thyroid patients and normal persons was collected from Kausar Hospital, PPHI (People Primary Health Invite) District Lab Khairpur and Civil Hospital Khairpur. To achieve the goal the data of 209 subjects were collected randomly. The serum samples were analyzed for lipid profile (total cholesterol, high density lipoproteins (HDL), low density lipoproteins (LDL), triglycerides (TGs) and very low density lipoproteins (VLDL) to evaluate dyslipidemia in thyroid patients. From 209 persons 98 were males and 111 were females. From thyroid patients 52.73% were hypothyroid, 27,27% were hyperthyroid and 20% were euthyroid. In hypothyroid patients prevalence of females was higher than males. This study concludes that hypothyroidism has impact on lipid profile levels i-e. HDL, LDL, VLDL and Cholesterol because in case of hypothyroidism their altered levels were observed.

## THE EFFECT OF FEEDING DIFFERENT LEVELS OF GREATER CELANDINE (CHELIDONIUM MAJUS) EXTRACT ON GROWTH PERFORMANCE AND DIGESTIBILITY OF BROILERS

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This study was conducted on 100-day-old broiler (*Gallus gallus domesticus*) chicks. The birds were distributed in to 4 groups (A, B, C and D) each group comprised of 25 chicks. The birds in A group was kept as control without supplementation of *chelidonium majus* extract, group B supplemented with 0.5ml/L of *chelidonium majus* extract, group C supplemented with 1ml/L of *chelidonium majus* extract and group D was sup-plemented with 1.5ml of *chelidonium majus* extract. The present study shows that maximum feed consumption (3405 g/b) was recorded in D group birds while in A group birds minimum feed consumption (3188) was recorded. In group C birds maximum water consumption and live body weight was recorded as (7487 ml/b) and (2100g/b) whereas in group A bird minimum water intake and live body weight was noted as (7013) and (3188 gm/b) respectively. maximum mortality and dressing

percentage was recorded as (65%) (5%) in B group birds, however in group C and A minimum dressing percentage (63) was noted and minimum mortality (3%) was noted in C group. Relatively better FCR (1.54) was noted in C group (where the birds were supplemented with 0.5ml/L of chelidonium majus extract) followed by group B (1.64), Group-D (1.66) and group A (1.72), respectively. Maximum relative weight of heart (0.54) was noted in group D, minimum weight of heart (0.41) was noted in group C. Maximum weight of liver (2.01) and spleen (0.12) were noted in group C, and minimum relative weight of liver (1.44) and spleen (0.11), were recorded in group B. Digestibility findings indicates that minimum crude protein (43%) was noted in A group birds while maximum increase of crude protein (61%) which was recorded in D group (A group known as control whereas, in D group 1.5ml/L of chelidonium majus extract was supplemented). Minimum crude fiber (54.53%) was recorded in A group while maximum crude fiber as well as dry matter was recorded as (61.67%) and (84%) in C group (A group birds remain control while birds of C group were supplemented with 1ml/L of chelidonium majus extract) but minimum dry matter (80%) was recorded in group A, B and D. Hence, economically from groups (A, B, C and D), the net profit remained Rs. 48.6, 46.8, 51 and 25.2/bird, respectively. Statistically, there was significant ( $p \le 0.05$ ) difference in feed intake among all groups (A, B, C and D), correspondingly. Significant (p≤0.05) higher feed intake was recorded in the birds of group D followed by B and C groups compared to the birds of group A. difference in weight gain group C showed significantly (p≤0.05) highest weight gain followed by birds in groups D and B compared to control group (A) birds. On dressing percentage there was non-significant (P≥0.05) difference between various treatment groups. There was non-significant (p≥0.05) difference in dressing percentage among all groups (A, B, C and D), correspondingly. The relative weight of heart was found significantly (P≤0.05) higher in the birds of group D followed by group C as compared to the birds of control group. Similarly, relative liver weight was significantly improved in birds of group C and D as compared with control birds. Likewise, our results further indicated that, relative weight of gizzard in the birds of group C was found to be significantly (P≤0.05) higher as compared with B and D as well as control groups. Furthermore, relative weight of spleen remained non-significant among all treatment groups. It was concluded that Better FCR was recorded for group C. Minimum mortality was also recorded in group C. The maximum digestibility of crude protein was observed in group D, but maximum digestibility of crude fibers and ash were recorded in group C. The broiler managed in group C proved to be more profitable as compared to rest of the treatment groups.

## DETECTION AND VALIDATION STUDIES OF TRACE METALS, PROTEIN AND STEROID IN DIFFERENT ORGANS OF LOCAL AND BRAND MEAT (POULTRY, CATTLE AND FISH)

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Present study was conducted to investigate the concentration of heavy metals (Na, K, Fe, Zn, Cu, Ni, Mg, Co, Cd and Pb) in different organs (heart, liver, muscles, wings, neck) of poultry, cattle (mutton, beef) and fish available in local and branded markets of Lahore. Atomic Absorption Spectroscopy and Flame Photometry were used to estimate the contamination of these metals in meat. The concentration of heavy metals in different organs showed great variation. The detected values of Na, K, Fe, Mg and Ni were under the tolerable levels cited by international standards; World Health Organization (WHO) and Australia New Zealand Food Authority (ANZFA). The Co, Cd and Pd were almost absent in red meat samples but poultry meat Home raised, Local, Menu and Zenith wing samples had high concentration of Cu exceeding the permissible limits. Rapid and sensitive Lowry method was adopted for scrutinizing the protein content in internal organs (heart, liver, breast, wings, neck and leg) of poultry meat (Home raised, Local, Zenith and Menu). The results showed that liver of Home raised chicken is a rich source of protein. Quantitative evaluation of steroid (testosterone) in chicken through High Performance Liquid Chromatography confirmed that the level of testosterone in different organs didn't exceed the acceptable limit.

#### A PROMISING ANTI-DIABETIC POTENTIAL OF DIDYMIN

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Didymin is a naturally occurring orally active flavonoid glycoside (isosakuranetin 7-O-rutinoside) and have several pharmacological activities. In this study, we evaluated the anti-diabetic potential of didymin via inhibition of α-glucosidase, protein tyrosine phosphatase 1B (PTP1B), rat lens aldose reductase (RLAR), human recombinant AR (HRAR), and advanced glycation end-product (AGE) formation inhibitory assays. Didymin strongly inhibited PTP1B, α-glucosidase, HRAR, RLAR, and AGE in the corresponding assays. Kinetic study revealed that didymin exhibited a mixed type inhibition against α-glucosidase and HRAR, while it competitively inhibited PTP1B and RLAR. Docking simulations of didymin demonstrated negative binding energies and close proximity to residues in the binding pocket of HRAR, RLAR, PTP1B and α-glucosidase, indicating that it has high affinity and tight binding capacity towards the active site of these enzymes. We also determined its molecular mechanisms underlying the antidiabetic effects of didymin in insulin-resistant HepG2 cells which significantly increased glucose uptake and decreased the expression of PTP1B in insulin-resistant HepG2 cells. In addition, didymin activated insulin receptor substrate (IRS)-1 by increasing phosphorylation at tyrosine 895 and enhanced the phosphorylations of phosphoinositide 3-kinase (PI3K), Akt, and glycogen synthasekinase-3(GSK-3), Interestingly, didymin reduced the expression of phosphoenolpyruvate carboxykinase and glucose 6-phosphatase, two key enzymes involved in the gluconeogenesis leading to a diminished glucose production. The results of the present study clearly demonstrated that didymin will be useful for developing multiple target-oriented therapeutic modalities for treatment of diabetes, and diabetes-associated complications.

### PREVALENCE OF HYPOTHYROIDISM IN PATIENTS WITH THYROID GOITRE AT NIMRA JAMSHORO, SINDH

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Jamshoro is Health and Education center of Sindh, along with densely populated area. Hypothyroidism is circumstance in which thyroid gland doesn't produce enough of certain crucial hormones. Thyroid disorder affects all ages and either gender. South Asian population has a particularly high prevalence of thyroid disorders mainly due to iodine deficiency. Its sole symptom is a visible goitre (enlargement of thyroid gland). Its true incidence is unknown though it is endemic in the northern areas of Pakistan especially in Swat, Dir and Chitral districts of Khyber Pakhtunkhwa and certain areas of Sindh province especially Jamshoro. In this context an attempt was made to study the Prevalence of Hypothyroidism in patients with thyroid goitre. The data was collected from 60 patients by questionnaire and bio chemical markers. It was noticed that the most common cause of goiter is iodine deficiency, malnutrition and poor living standard. Hypothyroidism is produces other several conditions, like 1 to 2% of the population has spontaneous hypothyroidism with 1.9% females and 0.1% males. It is more common with advancing age, affecting patients with age 40 or over. Women are affected 10 times more frequently than men. Severe hypothyroidism may lead to coma and death if untreated.

## BEHAVIORAL AND HEPATOPROTECTIVE EFFECT OF AQUEOUS EXTRACT OF CLOVE IN ETHONAL-INDUCED HEPATOXICITY IN ANIMAL MODEL RAT

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A methoxyphenol component of clove (eugenol) scientifically called as (Syzygium aromaticum) belong to (Family Myrtaceae) was suggested a lot of pharmacological effect which included antioxidant, anti-inflammatory, analgesic, antidepressant, anticonvulsant, antihyperglycemic, antibacterial, antifungal, hepatoprotective and antiviral effects. The aim of this study is to determine hepatoprotective activity of aqueous extract of clove. Total 18 rats were categorized into four groups. First group of 6 rats called as control(C), second group of 6 rats as model (M), and other 6 rats were used as test (T1). We induced chronic liver disease in test animals by giving 1 ml absolute ethanol orally daily for 9 weeks, no treatment was given to control and after inducing chronic liver disease 6 rats (T group) were treated with clove aqueous extract for 6 weeks. Along with treatment different behavioral experiments were taken to observe the effect of drug on animal. Statistical analysis was done using statistical package for social science (SPSS Version. 20). Clove exhibit stimulatory activity by increasing CNS activity. Open field activity showed that clove treated rat revealed motor activity increased. Clove treated rat gave anxiolytic effect in Light and Dark box spent more time in light as compare to dark. In the start week of experiment no such change were found in anxiety but in last week it reduced anxiety. It means this compound shown properties to cure anxiety rate. The liver function test of aqueous extract of clove treated rats showed less ALT, AST, ALP as compared to control which shown decrease in liver injuries, damages or liver disease. Hence it concludes that Clove aqueous extract shown significant effect on behavioral activity and also shown hepatoprotective effect on animal model rat.

## MORPHOMETRIC, PRODUCTIVE AND REPRODUCTIVE TRAITS IN INDIGENOUS CATTLE OF AZAD JAMMU & KASHMIR

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The morphometric study plays a vital role in the comprehension of the animal conformation. A total 237 female indigenous cows and heifers were studied from northern districts of Azad Jammu & Kashmir, with the objectives to describe the physical characteristics, reproductive and productive performance of indigenous cattle of AJ&K. Phenotypic characteristic were recorded visually while morpho-metric measurements were recorded by using measuring tape and vernier caliper. These cattle showed great variation in color. Five main colors (black 33%, red 20%, white 19%, brown 15% and grey 13%) were found in different parts of the body. The head length is 37.7±0.2 cm with small horns (11±0.3) cm). The positive correlation between Horn Length/Horn Width, Ear Length/Ear Width, Forehead Width/Horn Length were found statistically significant (p<0.01). The mean of body weight, heart girth, body length, height at withers, leg height, shin length, chest width, tail length, dewlap length, dewlap width, rump width, hump length, hump width udder depth, length of teat, distance between front teat, distance between rear teat, and body condition score (BCS) was 269.2±3.8 cm, 156.6±1.3 cm, 113.6±0.8 cm, 106.2±0.8 cm, 58.3±0.5 cm, 27.8±0.6 cm, 34.6±0.1 cm, 79.5±0.5 cm,  $51.9\pm0.4$  cm,  $13.7\pm2.8$  cm,  $29.0\pm0.4$  cm,  $22.8\pm0.2$  cm,  $17.7\pm0.2$  cm,  $20.9\pm0.4$  cm,  $2.3\pm0.1$  cm,  $3.9\pm0.0$  cm,  $3.2\pm0.1$  cm and 3.2±0.0 cm respectively. The highest correlations were 0.77 for Age/RW and 0.66 for Age/Parity. The average age at puberty was 40.96±0.5 months, mean±SEM age at first service was 51.29±0.9 months, mean±SEM age at first calving was 58.09±0.9 months, mean±SEM gestation period was 279.6±0.2 days, mean±SEM post-partum heat period was 253.2±14.8 days, mean±SEM service period was 257.69±5.8 days, mean±SEM calving interval was 17.10±0.2 months and mean±SEM service per conception was 1.5±0.06. The productive traits of the indigenous cattle were studied with milk yield per day 1.8±0.06litre. For an effective cross breeding the maintenance of pure parent lines must be attained with selection program for the improvement of genetics and specific characters of that line.

# PREVALENCE OF ANEMIA AND ASSOCIATED FACTORS AMONG PREGNANT WOMEN AT KAUSAR HOSPITAL MOTHER AND CHILD HEALTH CARE CENTRE KHAIRPUR MIR'S, SINDH, PAKISTAN

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Anemia is defined as a condition in which haemoglobin level is lower than the normal <11.0g/dl. According to WHO Anemia is an important problem affecting the developing countries including Pakistan. About one third of the global population (over 2 billion) are Anaemic. The aim of study was to determine the prevalence of Anemia in pregnant women of Khairpur city, District Khairpur Sindh, Pakistan. The cross-sectional study was conducted in Kausar Hospital Mother & Child Health Care Centre Khairpur, Sindh, Pakistan from August to December 2019. A total number of 100 pregnant women were included in this study randomly. A predesigned proforma/questionnaire was filled to obtain relevant information. The questionnaire was filled by face to face interview consisting of sociodemographic characteristics included Name, Age, gestational age, educational level, occupation and monthly income. Anemia was classified as per the World Health Organisation (WHO) criteria. Prevalence rate of Anemia in the present study was found to be 69%. The minimum Haemoglobin levels were observed 5.6g/dl and maximum Haemoglobin levels were observed 12.4g/dl with an average Haemoglobin level of 10.05 g/dL ±1.44. Among them 49% were with mild Anemia, 17% with moderate Anemia and 3% with severe Anemia. While the patients were divided into four age groups of 15-20 years, 21-25 years, 26-30 years and 31-35 years for the comparison of Anemia or Haemoglobin level. The minimum Age was observed 20 years and maximum Age were observed 35 years with an average age of 28.12 years ±2.5 years. The results showed higher prevalence of Anemia between age group of 26-30 years. It was observed that prevalence of Anemia was higher in 7-9 gestational months. The socio-economic factors (Age, Gestational month, Number of children and income source) showed significant association with Anemia. The study concluded that Anemia is common in pregnancy and is a chief public health issue in developing countries.

#### **EVOLUTION OF CEREBELLAR CORTEX IN VERTEBRATES**

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Cerebellum is a major component part of hind brain in vertebrates which is mainly involved in control of posture, balance, and coordination of voluntary motor activity. The present study aimed to study the comparative anatomy of cerebellar cortex of different vertebrates in order to develop inference for the anatomical transition during the course of evolution. Briefly, fifteen species of vertebrates including primates covering all major lineages of vertebrates were dissected to extract the whole brain to study several morphological features of brain for example length, mass and volume of full brain and its regions were taken and neutralized against body size. This was followed by histological investigation of the region where all three layers of cerebellar cortex namely molecular, purkinje and granular were explored in terms of cellular density and layer thickness. Statistical analysis and comparison were made depending on nature of data. Statistically significant differences in brain mass, brain length, brain volume, cerebellar length and volume was observed when each of the five major lineages of vertebrates fishes, amphibians, reptiles, birds and mammals were compared against each other. Negative correlation was found in total brain mass and volume, and cerebral length and volume against evolutionary time points suggesting increase in both the parameters from fishes to mammals. In contrast positive correlation of midbrain and cerebellar length and volume points to the reduction in anatomical span of both the regions over the period of time from fishes to mammals. Cellular density and thickness of both molecular and granular layer and interpurkinje distance was found significantly different in statistical terms between different lineages of vertebrates. However granular layer thickness was found positively correlated with the evolutionary time points which denotes reduction in layer thickness from fishes to mammals. The molecular layer thickness and inter purkinje distance remained unchanged over the period of time. Interestingly cellular density of both molecular and granular layers increases during transition from fishes to mammals. Our findings provide important matrices that have been altered during vertebrate evolution. This in turn reflects the sculpting forces that underscore the anatomy of vertebrate brain and cerebellum.

#### OLIVE LEAF EXTRACT TO ENHANCE THE FUNCTIONAL PROPERTIES OF FOOD

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Olive leaves are the waste of oil processing industry, which represents to be a good source of antioxidant and phenolic compounds as well as nutritional properties. An experimental analysis has shown the probability of enhancing the nutritional value of fermented table olives by the addition of olive leaf extract. Olive leaf extract when added to table olives and brought to ferment using yeast culture and native bacteria in the presence of a starter culture (*L. plantarum*) strain, the analysis has shown that olive leaf extract addition results in producing the fermented olives with higher level of antimicrobial, antioxidant and anti-inflammatory properties. Moreover, olive leaf extract and commercial starter act synergically against spoilage microorganisms. Thus, the olives fermented with olive leaf extract has quite a less bitter taste. Although the use of olive leaf extract is highly recommended when baked appetizers are produced with low quality extra virgin oil which therefore doesn't have a good content of antioxidants because olive leaf extract addition has excellent ability to reduce oxidative degradation of lipids. Antioxidants of olive leaf extract are stable towards heat with retaining 74.14, 67.03, and 75.59 % of its activity after boiling for 35 minutes at 100 degrees, dry heating for 30 minutes at 100 degrees, and autoclaving at 121 degrees for 30 minutes respectively. Olive leaf extract and hydroxytyrosol provides a cardioprotective effect by endoplasmic reticulum stress that prove to be a good source for the inhibition and cure of cardio vascular diseases. These fundamental properties highlight the scope of olive leaf extract for utilization in food and biological systems.

## DIET COMPOSITION OF MUSK DEER (MOSCHUS CHRYSOGASTER) IN ASTORE VALLEY, GILGIT BALTISTAN

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Musk deer (Moschus chrysogaster) are small solitary ungulates that inhabit the forested and alpine scrub habitats of mountains in Asia up to elevation of 2500 m. They are most active between dusk and dawn, alternately resting and feeding throughout this period. The Himalayan Musk deer is listed as critically endangered in Pakistan and listed under Appendix I of CITES. Present study was conducted to determine the diet composition of musk deer in Astore Valley, Gilgit-Baltistan and to find the similarity index between seasonal feeding of musk deer in the study area. Fecal samples of winter season were collected in October 2017, while the summer samples were collected in June 2018. Samples of 13 plant species including trees, shrubs and herbs were also collected simultaneously with the fecal samples form the study site based on the presence of the musk deer in the area. The samples were used to prepare reference slides for identification of plant species which was done through Micro-histological analysis. The slide observation was carried out in a systematic manner. Results revealed that a total of 13 plant species were recorded in the fecal samples during summer season while 11 plant species were recorded in the fecal samples during winter season. Distinct seasonal variation in the feeding patterns of Musk deer was observed in both seasons. The preference for trees (browse) (58.2%) is higher and more pronounced during winter and that change towards higher consumption of forbs (52%) during the summer season. However, the Musk deer continues to feed on both browse and forbs in both seasons. The diversity of herbs consumed by musk deer might suggest that it needs a variety of diet components hence calling for stronger conservation measures for herbs in its native habitat.

#### EFFECTS OF IODINE DEFICIENCY ON HUMAN HEALTH

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Iodine deficiency in life damage cognition and growth, but iodine status plays a key role in identification of thyroid disorders in adult age. Severe iodine deficit causes hypothyroidism and goiter because, despite a rise in thyroid. Iodine level is too low to much decrease the production of thyroid hormones. Iodine deficiency is chronic, endemic mental retardation, decreased fertility rate, endemic goiter and cretinism, increased perinatal death. Chronic thyroid stimulates results rise in the occurrence of toxic nodular goiter and hyperthyroidism in populations. The high prevalence of nodular autonomy causes rising the occurrence of hyperthyroidism if intake of iodine frequently increases by salt iodization. Lack of iodine origin not only goiter, but also mental deficiency, hearing loss and other short stature, neurological impairments due to thyroid is deficiency during fetal development and childhood. Although iodinated salt is available hypothetically in most countries where it is needed, its quality and share of the market are often insufficient. In many countries where only household salt is iodinated the iodine content has been set too low owing to an overestimation of household salt consumption. However, the government commended to pass regulation and provide means for efficient iodination of salts wherever this is important. Thus, optimization of population iodine intake is an important component of preventive health care to decrease the incidence of thyroid disorders. Iodinated salt should be used on daily basis in enough quantity, should has a constant and enough iodine content and is very economical as compared to other salt.

# ASSESSMENT OF BIOCHEMICAL AND OXIDATIVE STRESS RESPONSE IN POLYCYSTIC OVARIAN SYNDROME PATIENTS

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Polycystic Ovarian Syndrome considered as most communal endocrine abnormality during puberty in women which mainly caused by hormonal imbalance, excess androgen level and oxidative stress. Obesity, infertility, ovarian cancer and other menstrual disorders are major consequences of PCOS. Anti-infertility drugs i.e. Clomiphene Citrate is considered as first line treatment for infertility. To evaluate the effect of Antioxidant activity, hyperandrogenism, Clomiphene Citrate in PCOS patients. 5.0 ml venous blood sample of 60 patients of PCOS treated with Clomiphene Citrate and 60 Blood sample of Healthy individuals was taken in clotted gel vial from gynaecology department, Jinnah hospital Lahore. Blood was further processed for the estimation of Reduce Glutathione (GSH), Catalase (CAT), Superoxide Dismutase (SOD), Malondialdehyde (MDA), Estimation of Nitric oxide (NO), Estimation of micronutrients (Vitamin A, Vitamin C and Vitamin E), Estimation of AOPPs, estimation of AGEs and FSH. The serum NO level in diseased persons is  $1.48 \pm 0.11$  while in control persons is  $4.12 \pm 0.95$ . The serum NO is significant statistically (p=0.000 < 0.05). The serum FSH Level in diseased subjects' is  $14.65 \pm 32.60$  while in healthy subjects is  $19.34 \pm 22.40$ . The serum FSH is significant statistically (p=0.007 < 0.05). The serum Vitamin A Level in diseased persons is  $7.96 \pm 1.23$  while in healthy persons is  $5.21 \pm 1.00$ . The serum Vitamin A is significant statistically. (p=0.000 < 0.05). The Polycystic Ovarian Syndrome is very common issue throughout the world. In present study, a strong correlation exist between Antioxidant activity and biochemical response in PCOS patients receiving Clomiphene Citrate.

### 7. TOXICOLOGY

#### EFFECTS OF METALLOID SELENIUM ON DIFFERENT INSECT GROUPS

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Selenium is an essential nutrient for organisms. Supplements of selenium reduce cancer, heart diseases and HIV infection in humans. Their high doses cause direct toxic effects on insects and also affect their population indirectly via accumulation in plants or plant parts. Previous studies have reported negative effects of selenium on insect pollinators particularly in managed bees (*Apis mellifera* L). Its hyper-accumulation in plant cause high mortality in bees due to their pollen or nectar foraging. Moreover, high mortality in larvae of bees as well as sub-lethal effects on their development have been observed under laboratory conditions. Contrarily, hyper-accumulation of Selenium in plant did not affect the visitation of pollinators. Likewise pollinators, selenium is also reported to show anti-feedant effects against various insect pests i.e., aphid, western flower thrips and some orthopterans (crickets and grasshoppers). Selenium increases the stress tolerance in crop and provide defense against many herbivores pest. Therefore, selenium and related compounds can be integrated in IPM program of insect pests control; however its high dose may have negative effects on insect pollinators.

### EVALUATION OF CHROMIUM INDUCED GENTOXICITY IN BLOOD ERYTHROCYTES OF FISH, WALLAGO ATTU BY MICRONUCLEI ASSAY

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In this experiment geno-toxicity was estimated in peripheral blood erythrocytes of carnivorous fish, *Wallago attu* subjected to chromium toxicity by applying micronuclei test. Calculation of micronuclei (MN) and nuclear abnormalities (NAs) (deshaped and binucleated nuclei) frequency can reflect the degree of DNA damage. Results showed that the MN and NAs frequency was changed with durations and concentrations of chromium. The toxic potential of different concentration of Cr induced the genotoxicity in the following order: 1/3>1/4>1/5>1/7<sup>th</sup> of LC<sub>50</sub>. The nuclear abnormalities varied with duration as 28>21>14>7days. Statistical analysis showed a significant effect of concentration of Cr as well as duration of treatment on frequency of micro-nucleated erythrocytes.

### ASSESSMENT OF NUCLEAR ABNORMALITIES IN ERYTHROCYTES OF LABEO ROHITA EXPOSED TO ACUTE CONCENTRATION OF ENDOSULFAN+ CHLORPYRIFOS MIXTURE

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The experiment was conducted to evaluate the genotoxic potential of insecticides endosulfan+chlorpyrifos mixture on erythrocytes of fish, *Labeo rohita*. The fish was exposed to 96-hr LC<sub>50</sub> concentration  $(1.95\pm0.02~\mu g L^{-1})$  of chlorpyrifos+endosulfan mixture for 4-days. Blood was collected from caudal vein of fish after 1-day interval. The geno-toxicity was measured in term of micronuclei (MN) and other nuclear abnormalities (NAs) viz., binucleated nuclei, dumble nuclei, blebbed nuclei, notched nuclei and deshaped nuclei. Results concluded that the frequency of MN and NAs were significantly increased in erythrocytes of fish due to exposure of insecticides mixture in duration dependent manner as 1-<2-<3-<4-day.

### SIZE RELATED GROWTH PERFORMANCE OF CHANNA MARULIUS DURING CHRONIC STRESS OF WATER-BORNE COPPER

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The aim of this study was to investigate the growth performance of three length groups (50, 100 and 150mm) of carnivorous fish species (*Channa marulius*) under sub-lethal ( $1/3^{rd}$  of LC<sub>50</sub>) exposure of water-borne copper, in terms of increase or decrease in average wet weights (g) and total lengths (mm), condition factor, feed intake (g) and feed conversion efficiency (%). All the three length groups of *C. marulius* were exposed to water-borne copper, separately, with three replications, for 120 days, at controlled laboratory conditions. Weekly data on all the growth parameters of copper treated and control fish were collected and analyzed statistically. The results showed that treated fish species exhibited significantly lower average wet weight and total length increments than that of the control fish. Among three length groups of *C. marulius*, 150mm length group exhibited significantly higher average wet weight and total length increments than 100mm and 50mm length groups. The condition factor (K) values computed for 50mm length group of the fish were higher than 100mm and 150mm fish length groups. The copper treated fish species exhibited significantly lower feed intake than control fish. However, among three length groups of fish, the order of feed intake was 150mm > 100mm > 50mm. The FCE of three length groups of fish showed the order:  $50mm > 150mm \ge 100mm$ .

## EFFECTS OF GLUTATHIONE IN THE REGENERATION OF KIDNEY PARAMETERS AFTER ADMINISTRATION OF PHENOBARBITAL IN RATS

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To Examine effects of glutathione in regeneration of kidney parameters after administration of phenobarbital in rats. Investigational drug Glutathione was used as an anti-oxidant drug. Capsules of Glutathione (30mg) were dissolved in de-ionized water and 80mg/kg body weight was given to GLU group of female Albino Wistar rats; orally. Three behavioral studies were examined after one week of administration i.e. Open field experiment; particular for kinesis, light & dark experiment; specific for anxiety and home cage experiment; particular for stimulation. These experiments were scored for 5 minutes after the duration of a week for 2.5 months. For the determination of the level of regeneration of urea and creatinine, rats were decapitated and the blood sample was collected for the detection of kidney parameters' estimations by "RC-TECH (68000)" by Photometry technique. Results were carefully compared with control and model groups (untreated and phenobarbital treated groups respectively). Careful observation of results states, locomotion and stimulation in rats, that were gradually reduced after the phenobarbital administration, has steadily but significantly increased after treatment with GLU and the depression was markedly disappeared after the treatment. On Urea and Creatinine, Glutathione has showed better improvements when they were emphasized by comparing with control and model reports. Glutathione has constructive effects on locomotion and stimulation and decreases the level of anxiety in the body. These activities were depending upon the anti-oxidant concentrations. Glutathione showed better effects on Kidney parameters.

### PHYTOCHEMICAL, ANTIOXIDANT AND ANTICANCER ACTIVITIES OF OXALIS ACETOSELLA AND BASSIA INDICA KHYBER PAKHTUNKHWA, PAKISTAN

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Medicinal plants have been used for treatment of various disease from minor wound to chronic heart diseases worldwide. This study is aimed to find out the phytochemical composition, antioxidant activity and the anticancer activity of *B. indica* and *O. acetosella*. *Plant samples (B. indica* and *O. acetosella) were* from Khyber Pakhtunkhwa, Pakistan and were grinded. Ethanolic extract of both plants were checked for major phytochemicals, antioxidant and anticancer assay. Different concentration of ethanolic extract of *B. indica* and *O. acetosella* was used for anticancer activity against MCF7 (Brest cancer), HepG2 (Liver cancer), HeLa (Cervical cancer) cell lines for 48 hours. All cancer cells line viability lower with low doses of ethanolic extract of *B. indica* and *O. acetosella* in low dependent manner. Through the light microscope cells were morphologically examined and it indicated that after 48 hour ethanolic extract of *B. indica* and *O. acetosella* induced the morphological changes in colon cells. It is concluded that *B. indica* and *O. acetosella* ethanolic extract has anticancer activities against cancer lines. So, *B. indica* and *O. acetosella* extract and associated product may offer a new approach to the treatment and chemoprevention of Brest, Liver and Cervical cancer.

## STUDY ON GENOTOXIC EFFECTS OF BIFENTHRIN IN PERIPHERAL BLOOD ERYTHROCYTES OF CATLA CATLA USING COMET ASSAY

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Bifenthrin (Pyrethroids) belongs to the most commonly used pesticides throughout the world. Their extensive usage is a threat to the natural environments including aquatic ecosystems. Although bifenthrin are rapidly degraded in soil and plants, but they are extremely toxic to the fish. Keeping in view the high sensitivity of fish to bifenthrin, the present study was conducted to detect the genotoxic effects of bifenthrin in peripheral blood erythrocytes of freshwater fish, Catla catla using Comet assay. Fish were exposed to four sub-lethal concentrations viz. 10, 20, 33 and 50% LC<sub>50</sub> of bifenthrin along with positive (Cyclophosphamide) and negative control groups, separately, for 30 days. The DNA damage in terms of types of DNA damage (Type I – IV) from which percentage of DNA damaged cells, genetic damage index (GDI) and cumulative tail length of comets (µm) in peripheral erythrocytes of Catla catla exposed to different sub-lethal concentrations, with negative and positive control groups were also determined. The erythrocytes of Catla catla showed significantly variable effects of different concentrations of bifenthrin on percentage of DNA damaged cells, GDI and cumulative tail lengths of comets at p<0.05. The 50% LC<sub>50</sub> exposure of bifenthrin showed significantly higher DNA damage in terms of percentage of DNA damaged cells and GDI followed by that of positive control, 33%, 20%, 10% of LC<sub>50</sub> and negative control. However, cumulative tail lengths of comets were significantly maximum due to positive control followed by that of 50% and 33% LC<sub>50</sub> exposure of bifenthrin. Significantly positive concentration dependent increase in DNA damage was also observed during present investigation. This study also reveals that bifenthrin is very toxic to the fish and Comet assay can be used as useful tool for the determination of genotoxic effects of pesticides on fish.

## CORRELATION OF PEROXIDASE ACTIVITY WITH METAL BIOACCUMULATION IN FARMED AND RIVERINE CHANNA MARULIUS

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Aquatic environment is polluted by different pollutants including heavy metals which are frequently brought by increasing agricultural, domestic, industrial and commercial activities. For protection from these pollutants every aerobic organism possesses antioxidant defense system. Peroxidase is an important part of antioxidant systems which protects an organism from oxidative stress by reducing the harmful effects of reactive oxygen species (ROS). The present study was conducted to evaluate the influence of selected heavy metal pollution including Cr, Co, Cd and Ni on antioxidant enzyme activity in gills, liver, kidney and muscle tissues of farmed and riverine Channa marulius. The fish samples were collected from Trimmu Headworks of River Chenab with the help of gill nets. After collections of fish samples, the organs were extracted out by dissection at the sampling site. The extracted organs were preserved in ice boxes after keeping them in tagged polythene bags for transportation to the laboratory for further analysis. Water samples were also collected from sampling sites for the analysis of various physico-chemical parameters and for the detection of selected metal level. The extracted organs were divided into two parts in the laboratory. One part was for enzyme assay at 470 nm and the second part of extracted organs was digested on hot plate to determine the level of selected metals. For comparison, the farmed fish were sampled from semi intensive pond of fisheries research farms of the university. The peroxidase activity of extracted organs was calculated by atomic absorption spectrophotometer. Means ±SD was calculated and found that all the means are significantly different. The peroxidase activity in gills, kidney liver and muscles tissues of riverine fish samples was measured as 187.00±2.054Uml<sup>-1</sup>, 161.00±2.160Uml<sup>-1</sup>, 213.67±0.816Uml<sup>-1</sup> and 140.00±0.816Uml<sup>-1</sup>. While in farmed fish, was measured as 117.67±0.816Uml<sup>-1</sup>, 124.00±0.816Uml<sup>-1</sup>, 136.00±1.247Uml<sup>-1</sup> and 105.00±0.816Uml<sup>-1</sup> respectively. Overall the peroxidase activity in reverie fish was found to be higher as compared to farmed fish sample. The order of selected metals was observed as Cr>Ni>Co>Cd in riverine and farmed *C. marulius* gills, liver, kidney and muscles tissues, respectively. The inferences of the present study will be helpful in assessing the influence of metal pollution on the activity of antioxidant enzymes of fish.

# ASSESSMENT OF TOXICOLOGICAL EFFECTS OF HEAVY METALS (CADMIUM AND MERCURY) ON HEART AND PHARMACOLOGICAL INTERVENTION BY VITAMIN C IN RABBITS

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Mercury and cadmium are highly dangerous metals that can lead to disastrous effects in animals and humans. The aim of the current research was to elucidate the poisonous effects of mercuric chloride and cadmium chloride individually and in combination on biochemical profiles of plasma and their accumulation in heart. The therapeutic effect of vitamin C against these metals in rabbits was also studied. Mercuric chloride (1.2 μg/g), cadmium chloride  $(1.5 \mu g/g)$  and vitamin C (150  $\mu g/g$  of body weight) were orally given to treatment groups of the rabbits (1- control; 2-vitamin; 3- CdCl<sub>2</sub>; 4- HgCl<sub>2</sub>; 5- Vitamin + CdCl<sub>2</sub>; 6- vitamin+HgCl<sub>2</sub>; 7- CdCl<sub>2</sub>+HgCl<sub>2</sub> and 8vitamin+CdCl<sub>2</sub>+HgCl<sub>2</sub>. After the biometric determination of all intoxicated rabbits, biochemical parameters viz; low density lipoproteins (LDL), high density lipoproteins (HDL), cholesterol and creatine kinase were analyzed by using available kits. Levels of cholesterol (0.7±0.1 mmol/l), creatine kinase (2985.2±11 IU/L), LDL (20.35±1.31 mg/dl) were significantly (P<0.05) increased, HDL (84.78±4.30 mg/dl) was significantly (P<0.05) decreased while supplementation of vitamin C decreased the adverse effects of CdCl<sub>2</sub> and HgCl<sub>2</sub> on biochemical parameters in all metal exposed groups. Similar trend was also seen in rabbits treated with CdCl<sub>2</sub>+Vitamin and vitamin+CdCl<sub>2</sub>+HgCl<sub>2</sub>. Accumulation of Cd and Hg were higher in heart tissues. This study therefore, provides awareness on the toxicity of the mercury and cadmium chlorides, both alone and in their combination on a few biochemical parameters related to the cardiac functions in the rabbits and the possible protective role of vitamin C against the perturbations induced by metals is also elucidated.

### OCCURRENCE AND TOXICITY OF CADMIUM (CD) IN SEAFOOD ASSOCIATED WITH HUMAN HEALTH

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Shellfish are an important source of toxic metals, but also of essential elements in the diet. Seafood provides the long-chain omega-3 fatty acids, vitamins and minerals, which are essential to maintain good health. Moreover, seafood is a source of contaminants such as heavy metals and persistent organic pollutants that may affect health. Despite the existence of a legislation regarding seafood contaminants, food safety control in Pakistan is a matter of great concern. The aim of the present study was to determine that in what quantities seafood consumption would

provide nutritional benefits, while minimizing the risks linked to food contaminants. This study investigates the Cd level in commercially important seafood available in Karachi harbor and market. The aim of this study was to determine the impact of Cadmium contamination and accumulation in seafood and human health from the field and through Bioassay. The health risk associated with the Cd intake through seafood ingestion was evaluated. The results indicated that seafood consumption does not pose a significant health concern in the case of the usual consumption rate which considered typical for the human. However, a highly frequent consumption of seafood can have adverse health effects.

# TOXICOLOGICAL EFFECTS OF WATER-BORNE COPPER ON THE ANTIOXIDANT ENZYMES ACTIVITIES OF MAJOR CARPS

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Metals containing contaminated waters from industries and agriculture run off caused oxidative stress in aquatic fauna by enhancing the production of reactive oxygen species. To overcome the hazardous impacts of reactive oxygen species, there existed antioxidant enzymes defense system, in which peroxidase and catalase are the first lines of defense. Therefore, during present research work, effects of copper on peroxidase and catalase activities in two major carps viz. Labeo rohita and Catla catla were evaluated. 120-day old fish fingerlings of L. rohita and C. catla with three replications for each treatment, were exposed, separately, to 1/4th and 1/5th of their respective 96-hr LC<sub>50</sub> values of lead in glass aquaria for 14, 28, 42, 56, 70 and 84 days at controlled laboratory conditions along with control (untreated) fish group. After each 14-day exposure intervals, fish from all the treatments were sacrificed and their organs (liver, gills, kidney, brain and muscles) separated for peroxidase and catalase assays. Copper exposure induced significant variations in the activities of peroxidase and catalase in the selected organs of two fish species, L. rohita and C. catla as compared to control. Exposure to copper caused increased in peroxidase activity in L. rohita as compared to C. catla, while in all organs copper caused a significant increase in all organs as compared to control, but catalase enzyme showed a significant decrease in all the organs as compared to control. In both the fish species, peroxidase activity increased significantly with increasing exposure duration from 14 to 84 days due to enhanced production of reactive oxygen species whereas catalase showed decreased activity as increasing exposure duration to copper while among organs, liver showed maximum peroxidase and catalase activities followed by gills, kidney, brain and muscles, while among the fish species.

# PATTERNS OF HEAVY METAL ACCUMULATION IN DIFFERENT ORGANS OF BIGHEAD CARP (HYPOPHTHALMICHTHYS NOBILIS)

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Bioaccumulation of metallic substances in fish causes serious warning to the human after consumption. So, it is essential to find out accumulation pattern in fish. During current study, the accumulation patterns of metals in organs of bighead carp (*Hypophthalmichthys nobilis*) were determined after acute exposure. In order to calculate acute metal concentrations (LC<sub>50</sub> and lethal concentration) after 96 hour, bighead carp were subjected to different metal concentrations of Zinc and Iron, separately. Each metal concentration was tested in three replications. Bioaccumulation trends of treated metals (Iron and Zinc) were examined in selected fish organs such as muscles, gills, kidney and liver that were isolated after dissection for estimation of metal concentrations. The mean 96-hr LC<sub>50</sub>

of Iron and Zinc for *Hypophthalmichthys nobilis* was calculated as  $123.3\pm0.79~\text{mgL}^{-1}$ ,  $48.28\pm0.82~\text{mgL}^{-1}$  while lethal concentration was  $175.42\pm3.30~\text{mgL}^{-1}$ ,  $84.08\pm1.27~\text{mgL}^{-1}$  respectively, determined by probit analysis method. The metal accumulation pattern for Iron at 96-hr LC<sub>50</sub> concentration and lethal concentration was same and recorded in order as followed: Kidney > gills > liver > muscles. The metal accumulation pattern for Zinc at 96-hr LC<sub>50</sub> concentration and lethal concentration was noted as followed: Liver > gills > kidney > muscles. Results concluded that bioaccumulation patterns of metallic substances in fishes essential to be done for collection of helpful information for the evaluation of the possible health risks of metals.

## THE EFFECT OF ACUTE EXPOSURE OF NI AND ZN ON THE BEHAVIORAL RESPONSES AND HAEMATOLOGICAL PARAMETERS OF BIGHEAD CARP

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Water pollution poses the major threat to human life and other aquatic organisms. Heavy metals are the main cause of contamination which causes adverse effects on fish. In order to determine the lethal effects of heavy metals like Ni and Zn, study was conducted on bighead carp. Glass aquaria of 70 liter water holding capability were used for experiment, each containing 12 juvenile of bighead carp. The experiment was performed in triplicate. The bighead carp were exposed to zinc and nickel to LC<sub>50</sub> concentration during acute toxicity test. Under the exposure of treated metals, the behavioral responses and hematological parameter i.e. erythrocytes, leucocytes, haemoglobin (Hb), haematocrit, mean corpuscular volume (MCV), mean corpuscular haemoglobin and mean corpuscular haemaglobin concentration (MCHC) were recorded. Maximum reduction in hemoglobin contents, hematocrit, red blood cells were observed in treated groups then that of control group. The values of mean corpuscular hemoglobin concentration, mean corpuscular volume and mean corpuscular hemoglobin presented significant decrease in both nickel and zinc treatments. Number of leucocytes increased when fish exposed to zinc and decreased after nickel exposure. The bighead carp also showed the abnormal behavior e.g. swimming very fast, jerky movements, faster opercular beating, gulping of air and loss of equilibrium after 96 hours. Analysis of variance was used to compare variables among both control and treated fish. Correlation was used to determine the relationship between different variables.

### ACTIVITY OF ANTIOXIDANT ENZYMES AS INFLUENCED BY METAL BIOACCUMULATION IN VARIOUS ORGANS OF CATLA CATLA FROM RIVER RAVI

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Antioxidants are considered as important biomarkers in evaluating the health of aquatic system. Two essential antioxidant enzymes, catalase and peroxidase use a novel therapeutic way to overcome oxidative stress. They show dual nature in battling against metal ions comprising not only scavenge but also chelating with active sites of ROS. The present research is designed to determine the antioxidant enzymes activity as influenced by metal bioaccumulation in various organs of *Catla catla* from three sites of River Ravi. For this purpose, various fish organs (liver, kidney, gills, cardiac and muscle tissues) were extracted out and placed in tagged polyethylene bags, preserved in ice container and brought to laboratory for further analysis. Extracted organs were processed through standard methods for assay of metal bioaccumulation and enzyme activity. Activity of catalase and peroxidase was determined by using UV-Visible Spectrophotometer at 240nm and 470nm, respectively. Metal bioaccumulation was assessed by using Atomic Absorption Spectrophotometer (AAS). This study showed elevated antioxidant enzymes activity in various organs of riverine *C. catla* than pond cultured fish. The sequence of antioxidant activity in fish organs

sampled from River Ravi was following: liver>kidney>gills>heart>muscles. The bioaccumulation of metals (Cu, Pb) among three sampling sites was found in the following order: Baloki Headworks>Shahdara Bridge>Sidhnai Headworks. Level of lead was found greater than copper in all studied sites of River Ravi. Bioaccumulation of metals was found maximum for liver following kidney, gills, heart and least for muscle tissues. Analysis of water samples showed significantly different physico-chemical parameters for all studied sites of River Ravi. Highly significant results (p<0.05) were obtained among all sampling sites for raised antioxidant activity as induced by metal ions. This research will be helpful in determining the extent of metal pollution in River Ravi, metal bioaccumulation in fish tissues and their impact on dynamics of antioxidant enzymes in fish.

### ASSESSMENT OF ORGAN SPECIFIC CATALASE ACTIVITY IN NILE TILAPIA (OREOCHROMIS NILOTICUS) FROM DIFFERENT POPULATIONS OF RIVER CHENAB

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Contamination of water bodies due to anthropogenic activities is the major issue at the present time. Such contaminants decompose vital molecules of body by causing oxidative stress and ultimately enhanced the mortality rate. The present study was accomplished to assess the catalase (CAT) activity in association with heavy metals in various tissues (gills, heart, hepatic, muscles and kidney) of Nile Tilapia (*Oreochromis niloticus*). For this purpose, samples of proposed fish and water were collected from selected sites of River Chenab. Organs were extracted out at the fishing sites and CAT activity was analyzed in the laboratory after transportation of samples preserved in labeled polythene bags in dry ice boxes. Extracted organs were divided into two equal halves, one part for metal assay and other for enzyme assay. Catalase assay was carried out at 240nm by using spectrophotometer. Water samples were assayed for the analysis of physico-chemical parameters. The experimented data was statistically analyzed at *p*<0.05 by using R program. The inference of the present research work showed higher catalase response towards oxidative stress in riverine tilapia than farm-raised *O. niloticus*. Sequence of catalase activity in various studied organs was as liver>gills>kidney>muscles>heart. Higher bioaccumulation of Cu and Cr in liver as well as in heart, Pb and Cr in gills was observed following the sequence Cu>Ni>Cr>Pb, Cr>Cu>Ni>Pb and Cr>Pb>Cu>Ni respectively. The inference of the study helped in estimation of antioxidant responses against oxidative stress produced by metal pollutants.

# HEAVY METALS VIZ. ZN, NI AND CU LEVELS IN SEDIMENTS AND WATER FROM BALOKI HEADWORKS TO SIDHNAI BARRAGE IN THE RIVER RAVI, PAKISTAN

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Heavy metals are non-biodegradeable and are naturally found in our universe. This study was designed to evaluate the toxicity levels of Zn, Ni and Cu in river water and sediments. Three main sites of river Ravi, Baloki headworks, Kamalia-Chichawatni bridge and Sidhnai barrage were selected to collect the samples of water and sediments. This research continued for six months (October, 2018 to March, 2019). Each studied site divided into further three sub-sites and samples collected on monthly basis. The collected samples of water and sediments were wet digested and then analyzed by Atomic Absorption Spectrophotometer. Differences in three metal's levels in water and sediments found to be in the following sequence: Zn > Ni > Cu. Maximum toxicity in water and sediments observed at Kamalia-Chichawatni bridge and the variations between sites were in following order: Kamalia-

Chichawatni bridge >Baloki headworks >Sidhnai barrage. The increase in toxicity was due to release of various chemicals and heavy metals in river from industries and domestic wastewater. The conclusions from this study suggested that at all studied locations, toxicity levels of metals in sediments were greater than that of water. Water temperature and pH had strong influence on the concentration of metals in water.

### EVALUATION OF SERUM BIOCHEMICAL PARAMETERS OF CIRRHINUS MRIGALA UNDER ACUTE EXPOSURE OF ARSENIC

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Arsenic is one of the most important pollutants in the aquatic ecosystem which has the adverse effects in fish. The objective of present study was to determine the acute toxicity of arsenic trioxide on serum biochemical parameters of *Cirrhinus mrigala*, at different exposure durations such as 24h, 48h, 72h and 96h. Different serum biochemical parameters such Na<sup>+</sup>, K<sup>+</sup>, Cl<sup>-</sup>, urea, total protein , total glucose , albumin , ALT and AST were analyzed in control and arsenic exposed groups. There were significant alterations in all the serum biochemical parameters under acute exposure of arsenic trioxide. Among serum biochemical parameters, the level of Na<sup>+</sup>, Cl-and albumin noticed lower at all exposure durations of arsenic, however the level of K<sup>+</sup>, total protein , urea , total glucose ,AST and ALT were noticed higher in arsenic exposed fish. The results indicated that arsenic trioxide has devastating effect on serum biochemical parameters and these serum biochemical parameters can be used as bioindicators of toxicity caused by arsenic trioxide.

# ESTIMATION OF FAUNAL DIVERSITY EFFECTED BY HEAVY METAL, CADMIUM AND LEAD CONTAMINATION IN RICE CROP IRRIGATED WITH TANNERIES WASTEWATER IN DISTRICT SIALKOT

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Sialkot, an agricultural as well as industrial city of Pakistan, specifically known for its leather industry is earning valuable foreign exchange per annum through its leather export. Although leather industry is providing employment to thousands of people, but tannery wastes discharged near agricultural land are affecting quality of environment including flora and related fauna. Rice crop is one of the rich ecosystem for insect diversity as it contains both arid and aquatic ecosystem. Present study is aimed at finding the effect of heavy metals cadmium and lead discharged from tanneries on water and soil quality which is directly linked with rice field and their associated insect fauna. Water, soil, plant and insect samples were subjected to analysis through atomic absorption spectroscopy for heavy metals detection. Insect collection was carried out by following sweep method. Data analyzed by using t-test and Shannon diversity index shows that there is relatively significant difference p<0.05 in insect diversity of both areas. Moreover, amount of Cadmium and lead accumulation is higher than permissible limits of NEQS, Pakistan in water, soil and insects associated with nearby rice crops of tannery area than those of non-tannery area. Hence, findings of the study will contribute in monitoring the tannery effluent disposal sites with respects to disposal planning of the tannery wastes. The information may offer basis for the future biological research on the bioaccumulation mechanism of insects.

### STUDY OF EFFECTS OF SELECTED METALS CADMIUM AND LEAD ON INSECT FAUNA OF BRASSICA CROP IRRIGATED WITH TANNERIES WASTEWATER IN DISTRICT SIALKOT

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In the 21st century rapid industrialization and urbanization has drastically increased the amount of discharged wastewaters accompanied with lethal chemicals. Leather industry has a major contribution towards the economy of Pakistan. The present study is designed to assess the diversity of insects associated with Brassica crop in district Sialkot and determination of heavy metals, lead and cadmium in soil, water, plant and insects samples. Sialkot is well known for its agricultural land and leather industries. Different types of wastes are generated by leather tanneries depending on the process used for tanning purpose, which create significant environmental challenges. This study deals with the investigation of physiochemical parameters of tannery and non-tannery areas soil and water. Heavy metals are known for their adverse effects on environment. Heavy metals including cadmium and lead were significantly analyzed in soil, water, insects and plants samples associated with tanneries. Atomic absorption spectroscopy was applied to investigate the concentration of Cadmium and Lead. The levels of cadmium and lead were above the National Environmental Quality Standards of Pakistan. Five months sampling was carried out to analyze the diversity of insect fauna associated with Brassica crop. Quadrat method was followed for insects collection and results showed at least 17% decline in tannery area insect fauna. Insects play a significant and vital role in normal functioning of ecosystem, and they are indicative of the environmental health. It is recommended that practice of installation of wastewater treatment units with tanneries should be established generally.

#### NEUTRALIZATION OF LEAD-INDUCED NEPHROTOXICITY IN ALBINO RATS BY EGB 761

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EGb 761 is standardized extract of the world's most ancient tree Ginkgo biloba and proposed for its efficacy against lead induced nephrotoxicity. It has also shown anti-oxidant effects in various human and animal studies. Our study aims to investigate the remedial effects of EGb 761 on lead-induced nephrotoxicity in albino rats. To monitor the effect of EGb 761 on microscopic anatomy and laminin expression in the kidneys of albino rats treated with lead acetate. Four groups of Wistar albino rats were recruited where one was kept as control, one was treated daily with 100mg/kg body weight of EGb 761 orally, one with 8 mg/kg body weight of lead acetate intraperitoneally daily and one treated daily with both lead acetate and EGb 761 in the same doses. The body weight of the animals was continuously monitored till the end of the experiment (42 days). At the end of the experiment the animals were sacrificed, kidneys retrieved, fixed and then subjected to the histological and immunohistochemical examination. There was significant drop in the body weight of rats treated with lead acetate as compared to the control group (p<0.0001). Deterioration of the microscopic anatomy of kidneys entailed by lead treatment was reasonably reversed by the inclusion of EGB-761 along with lead, as evident by the improvement in nuclear count (p=0.0169), tubular diameter (p=0.0027), nuclear diameter (p=0.0011) and urinary space (p=0.0202). In addition, expression of laminin in the basement membrane was improved in lead+EGB-761 treated rats as compared to the lead-only treated rats (p=0.0197).

## ACUTE INTOXICATION OF METALS IN *LABEO ROHITA* WITH SPECIAL REFERENCE TO THE PHYSIOLOGICAL AND BIOCHEMICAL EFFECTS

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An experiment to assess the changes in hematology and serum biochemistry in *Labeo rohita* exposed to metals was conducted. Results showed that the acute exposure of metals to fish caused significant variability (p<0.01) in hematological and serum biochemical parameters as compared to control. Copper exposure to the fish had more pronounced effects as it resulted in significantly (p<0.01) lower red blood cells count, hemoglobin content, hematocrit content and higher white blood cells count, while zinc exposure showed least toxic effects towards hematological parameters as compared to other metals. Among all the exposure durations of metals, the 96-hr exposure caused maximum negative effects on fish. Lower level of serum sodium, chloride, albumin and total protein was observed in fish under the exposure of copper as compared to other metals while potassium, aspartate aminotransferase and alanine aminotransferase levels were higher. However, the least toxic effect on all above mentioned biochemical parameters were noticed in zinc exposed fish. This study proposed that the occurrence of toxic metals in the aquatic environment has a strong impact on hematology and serum biochemistry of the fish.

### 8. VIROLOGY

## EPIDEMIOLOGY OF HEPATITIS C VIRUS INFECTION IN IDPs OF WAR AGAINST TERRORISM IN NORTH WAZIRISTAN AGENCY, PAKISTAN

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HCV infection appears to be endemic in most part of the world, with a prevalence of around 3% world-wide. HCV infection is life astounding health problems worldwide, with over 170–200 million infected people including about 17 million from Pakistan and it causes cirrhosis and other complications that often lead to death. However, our knowledge about the disease and its mechanisms is limited. The study was conducted in IDPs of North Waziristan Agency (NWA). A total of 1837 individuals were sampled including males and females and the age of 1–60 years of age and some samples from over 60 years aged individuals and 516 individuals (28.08%) were screened out as positive. They were categorized into 5 age groups. They were screened for HCV by rapid method (ICT). The prevalence rate of HCV infection is directly related with age *i.e.* higher the age group has the higher rate. Individuals with the risk factor of dental surgery also have the high prevalence.

# MOLECULAR EPIDEMIOLOGY OF MEASLES VIRUS GENOTYPES AND CLADE TYPES IN PESHAWAR, PAKISTAN

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Measles virus infection causes measles which is a highly contagious disease and is responsible for a large number of childhood mortality worldwide. This illness starts with fever, cough coryza, conjunctivitis and maculopapular rash. Globally about 40 million deaths occur due to measles and in only in Asia about 777,000 deaths occur every year. Measles virus (Paramyxoviridae family) has 8 clade types and 24 genotypes and has only one serotype. In Pakistan measles outbreaks occur regularly at different intervals in which genotypes and clade types are still not detected. Therefore in this study through multiplex PCR the genotypes are clade types are detected instead of using sequencing for enhancing measles control especially in developing countries like Pakistan. Throat swaps were collected from 100 patients in 2018-19 outbreaks from Peshawar. RNA was then extracted from these samples through trizol kit and then complementary DNA was formed and amplified through nested PCR A conventional genotyping method based on multiplex PCR was used to discriminate between all active circulating measles virus clades and genotypes using target specific primers. This study found different clades and genotypes of measles virus in which the most prevalent clade type is B (28%) and least one is clade G (4%). The most prevalent genotype founded is H<sub>2</sub> (48%) followed by D6 (41%), B<sub>3.1</sub> (37%), G<sub>2</sub> (37%) and D7 (35.5%). Other genotypes detected are B<sub>3.2</sub> (14%), D3 (28%), D4 (8%), G<sub>3</sub> (21%) and H<sub>1</sub> (8%). This study concluded that different genotypes are circulating in outbreaks in Pakistan and the vaccine must be needed to improve for the total eradication of measles from here. Also a large scale comprehensive study is needed to further explore the clade types and genotypes of measles.

# MOLECULAR AND HEMATOLOGICAL FINDINGS OF *DEN-3* AMONG CHILDREN IN LAHORE, PAKISTAN

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Dengue virus belongs to family Flaviviridae which is transmitted by mosquito i.e. Aedes egypti. During the last decade the cases of Dengue virus have been increased rapidly in children and death rate of more than two million has

been reported. The present study was conducted to find out the prevalence of genotypes of *Dengue virus* in children under the age of 15 years in Lahore, Pakistan and study the correlation of hematological features with the clinical findings. A total of 112 serum samples were collected from clinically suspected patients of Dengue fever from March, 2017 to December, 2018 from different tertiary care Hospitals at Lahore, Pakistan. Based on the age of the patients the samples were divided into four groups from A to D i.e. 0-1 year, 1-5 year, 5-10 year and 10-15 years of age. Rapid Immuno-chromatography (ICT) test was performed on collected serum samples followed by Quantitative Real-time PCR for serotype of *Dengue virus*. Out of 112 samples, 34 samples were DEN virus positive by rapid ICT screening method. No virus was detected in group A & B while 3 samples were positive in group C (1 boy & 2 girls) and in group D, 31 samples (23 boys & 8 girls) were positive. The results of RT-PCR showed DEN-3 serotype exclusively in all the ICT positive samples. On hematological analysis, elevation in hematocrits was noted in 41% dengue positive cases, leucopenia was prominent in 79% while thrombocytopenia was noted in 69%. Biochemical analysis indicated that liver enzyme were also raised in patients (ALT 89%, AST 79%) while lower level of cholesterol (69%,) and serum albumin (25%) were also observed. The results indicate that, the prevalence of DEN-3 serotype in children was hundred percent which indicated that DEN-3 serotype may cause severe epidemics in future in Lahore, Pakistan.

# IDENTIFICATION AND CHARACTERIZATION OF DEFORMED WING VIRUS IN HONEY BEE APIS MLLIFERA POPULATION OF KHYBER PAKHTUNKHWA PAKISTAN

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European honey bee (Apis mellifera) reasonably an important insect, provides crop pollination services and precious hive products, thereby increasing the value of global agricultural production by billions of dollars. The worldwide population of A. mellifera is under pressure from habitat loss, environmental stress and pathogens, particularly viruses that cause lethal effects. Deformed wing virus (DWV) is an important contributor to honey bee colony losses. DWV (family Iflaviridae) together with its vector mite (Varroa destructor), is likely the major threat to the worldwide A. mellifera population. It usually causes covert infections but can have devastating effects on bees health when transmitted through the ectoparasitic mite V. destructor, which is an external parasite that attacks and feeds upon honey bees. Recently in Pakistan, honey bee colony losses at an alarming rate have been reported, however, there remains no clear explanation for these colony losses, due to parasitic mites and viruses etc. In order to investigate DWV, the precise number of infected DWV adult and larvae of bees were collected. Out of the total 970 colonies, 580 symptomatic samples that include 182 (18.76%) adult deformed winged, 114 (11.75%) shortened rounded abdomen, 66 (6.80%) larvae and 218 (22.47%) mites were also observed. RNA extraction followed by using specific RT-PCR protocols for the detection of DWV accompanied by sequencing of the PCR products we demonstrate that the honey bees were indeed infected with DWV in Pakistan. The outcome of this study is to make awareness among beekeepers about this viral infection as well as its control in honey bees, and proper apiary management for the monitoring of DWV.

#### EVOLUTION OF HEPATITIS B VIRUS ONCOGENE, HBX IN PRIMATES AND RODENTS

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Hepatitis B Virus (HBV) is one of the leading causes of viral associated Hepatocellular Carcinoma. To date one oncogene, HBx is known for the virus encoding a 154 amino acid long protein. In this study we have modelled full

length molecular structure of the HBx by employing strategies of iterative threading approach followed by thermodynamic and structural refinements. In addition, phosphorylation potential of the protein was also explore on the basis of both primary and tertiary structures. The findings were then aligned with the orthologous sequences of HBx of different HBV genotypes and HBV of other primates and rodents. Sequence alignment showed that p53 binding regions are highly conserved amongst orthologous sequences compared to regulatory domain of HBx. Xtal pred analyses predicted low probability of crystallization of HBx thereby difficult to be structurally resolved empirically. The modelled structure showed extensive intrinsically disordered region at the N-terminal of the protein whereas the C-terminal is alpha helical in shape. Structural comparison of the full-length model of HBx of different HBV genotypes of humans and other animals showed subtle variation in the Cα back bone architecture where RMSD was found to be from 0.04Å to 0.68Å. Ser 25, Ser41 and Ser43 showed significant potential of being phosphorylated amongst different HBx orthologues. Consistently the most commonly kinase predicted to undertake HBx phosphorylation was found to be Unsp. In summary, the findings will provide insights to the evolution of HBx and its neo-functionalization that leads to the evolution of its oncogenic attributes.

# ABSTRACTS PRESENTED AT THE FIRST VIRTUAL CONGRESS OF ZOOLOGY

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### **SECTION - I**

### 1. HERBAL MEDICINE, BIOCHEMISTRY, BIOTECHNOLOGY AND BIOINFORMATICS

## EFFECT OF SOLANUM NIGRUM ON BLOOD BIOCHEMISTRY, CHOLESTEROL, GLUCOSE AND UREA LEVEL OF ROTENONE INDUCED PARKINSON'S RAT MODEL

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Parkinson disease (PD) is a most chronic andmost progressive disease of the central nervous system. On a daily basis more than 100 people are diagnosed with Parkinson's disease. Rotenone is an active compound of pesticide which is main cause of Parkinson's disease in modern age. To study the effect of Solanum nigrum polyphenols on Parkinson's disease rotenone induced Parkinson's model was prepared. *Solanum nigrum* fruit extract contents were identified by Gas chromatograph mass spectrometry (GCMS) analysis. Rotenone was administrated on a daily basis systemically by intraperitoneal injection of dose: 1.5 mg/kg, over a period of 28 days. Rats were divided into four groups. First group as control group given sun flower oil, second group was given rotenone third group was given rotenone + *Solanum nigrum* fruit extract and the fourth group was treated with *Solanum nigrum* fruit extract. The second group showed Parkinson disease which was confirmed by different behavioral and chemical analysis. The thirdand fourth group showed therapeutic effect as compared to rotenone treated group. Data analysis indicated that rotenone treated rats are capable of causing degeneration of dopaminergic neurons and induction of parkinsonian symptoms. Hemoglobin, platelets count, red blood cells in whole blood and in serum analysis blood glucose level, cholesterol level, triglycerides urea and creatinine were analyzed by all groups. The level of cholesterol and glucose in *solanum nigrum* treated group decreasedas compared to control and rotenone treatedgroups.

## ASSESSMENT OF SYNERGISTIC ANTIBACTERIAL ACTIVITY AND ANTICANCER EFFECT OF FIVE INDIGENOUS PLANTS

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Insufficiency of novel antimicrobial and anticancer compounds and increased resistance of cancer cells against drugs has compelled the researchers to find and develop the new therapeutic agents that could be used efficiently as alternatives to synthetic drugs or may help these drugs in combating the infections. The present study was conducted to assess the anti-cancer and antibacterial synergistic effect of five medicinal plants *Jacaranda mimosifolia*, *Bismarkanobilis*, *Cupheahookeriana*, *Aphelandra simplex*and *Choiysaternate*. Methanolic extracts of these plants were checked for the synergism against isolates of *Styphylococcusaureus* (2) and *Salmonella typhi* (1)through well diffusion assay.Cytotoxicity of the plant extracts was determined through MTT assay against HepG2 and HeLa cell lines.Phytochemicals of *J. mimosifolia*were also docked against Bcl-2 (anti-apoptotic protein) in order to find the appropriate inhibitor of this anti-apoptotic protein. Only one plant, *C.hookeriana* showed the most significant cytotoxicity against HepG2 cell line. While three plants *A. simplex*, *J. mimosifolia* and *C. hookeriana* showed significant cytotoxicity against HeLa cell lines. Out of fifteen compounds only two compounds 2,4-bis(1,1-dimethylethyl) phenol and 1-β-D-Ribofuranosyl-3[5-tetraazolyl]-1,2,4-triazole were identified as potential phytochemicals with strong binding capabilitesand efficient drug-like properties. After further research on these

newly identified cytotoxic plants, these plants and their bioactive compounds may be used for the preparation of efficient, safe and cost effective anti-cancer and antibacterial drugs.

# BIO-PHARMACEUTICAL EVALUATION OF TRIGONELLA FOENUM-GRAECUM L., NIGELLA SATIVA L. AND ZINGIBER OFFICINALE AGAINST BOVINE MASTITOGENS

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Livestock sector plays an important role in any nation's economy. Mastitis is one of the diseases that effects the livestock economy very badly. Bacterial mastitogens cause sensitivity that leads toward inflammation and tenderness of the mammary glands. Mastitis is usually cured by antibiotics, but due to development of resistance against antibiotic agents and these drugs are becoming ineffective against bovine mastitogens. Therefore, medicinal plants are being considered as an alternative source of cure. In present study three medicinal plants namely, Trigonella foenum-graecum (Fenugreek and methidane), Nigella sativa (Kalonji) and Zingiber officinale (Ginger) were selected for extract formation in n-Hexane, Acetone and Ethanol solvents. Milk samples from infected cows were collected from veterinary hospital and three strains of bacteria (Staphylococcus aureus, Escherichia coli and Klebsiella pneumonia) were identified and isolated, 24 different extract preparation were made (individual as well as combination) and applied against isolated bacterial strains. By using Disc Diffusion method zones of inhibition were measured. Results were compared with the standard antibiotic; Ciprofloxacin. Efficacy of extracts and combinations were also compared with each other. One way Anova test was used to determine the level of significance of our results. From all single extract, N. sat (n-hexane) showed maximum inhibitory effect (IZ=14.5 mm) with colonial growth inside the zone. Among the combination of two extracts N. sat: Z. off 1:1 (n-hexane) showed highest inhibitory effect (IZ=21 mm) against Klebsiella pneumonia. With combination of three extracts Z. off: N. sativa: T. foe (1:1:1) (n-hexane) showed highest potency against Staphylococcus aureus (IZ=14.5 mm). However, when all three extracts of single plant were mixed, N. sat (Ethanol: nhexane: Acetone) 1:1:1 gave best results against Staphylococcus aureus (IZ=20 mm) Ciprofloxacin showed potential only towards S. aureus. Results proved that from all three plants Nigella sativa and from all three extracts n- hexane is most effective agent to control growth of strains. Also, the combination of two or three extracts give better results as compared to single extract. Therefore, by formulation of various combinations of extracts multidrug resistant bacteria could be controlled. IZ = Inhibition zone.

#### INSECTS AS SOURCE OF PROTEIN IN POULTRY FEED; A REVIEW

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The demand of Poultry meat and eggs is increasing day by day and will increase in future. Poultry Feed is an essential component of poultry farm in poultry business. It is much expensive for small grower as well as at Commercial level. Protein source is costly that makes poultry feed expensive. About 20% to 23% protein is recommended in poultry feed depends upon energy required. Insects are the cheap and alternative source of protein with amino acids. By dry weight he averages insects contain 50% protein but some insects also contain about 75% protein. Silkworms, Locust, Black soldier Fly, Maggots, crickets, and grasshoppers can be safely fed to chickens without affecting the quality and taste of the meat. Insect based feed increases the nutritional value of poultry meat. The method of feeding insects to poultry will facilitate the development of agriculture-based recycling systems, reduce waste, and can also help to reduce environmental pollution. Production of Chicken meat is 1245 million kg annually in Pakistan and per capita consumption of Chicken meat is 6.22 kg and 56 eggs annually. Developed countries consume around 40 kg of Chicken meat and more than 300 eggs per capita every year. It is possible to increase per capita of both poultry meat and eggs per year in Pakistan. Different methods are used in insects rearing depends upon the life stage. At larval stage, they are rear in boxes. The box contains several shelves to minimize the

production space of each insect. In some cases, stackable boxes are used and placed on trucks or pallets to allow free movement in the rearing area. These methods are used to prevent any toxic material that can cause economical loss. The aim of this paper is to provide cheap and easy source of protein with amino acids for Poultry feed and increase production.

### CHARACTERIZATION OF FREE LIVING AMOEBAE FROM DRINKING WATER DISTRIBUTION SYSTEM AND ITS CONTROL USING DISINFECTANTS

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Free-living amoebae (FLA) are ubiquitous unicellular protozoans that are pathogenic or reservoirs of pathogenic bacteria and viruses, causing serious human and animal infection. The present study revealed occurrence, isolation, morphological characterization, and disinfection treatment of FLA from drinking water resources of Lahore, Pakistan. A total 150 drinking water samples were taken from different areas of Lahore. Acanthamoeba with acanthopodial morphotype was observed in three samples, four samples were positive for monopodial morphotype of Hartmannella while Allovhalkampfia was observed in two samples. Acanthamoeba was characterized by flat-shaped trophozoites (25-30 µm) having acanthopodia and they form double-walled cysts of 10-20µm. Hartmannella was characterized by uninucleated cylindrical trophozoites and rounded, single-walled cysts (11-16 µm). The trophozoites of Allovahlkampfia had eruptive elongated monopodia. The maximum number of trophozoites were observed after the 5th of inoculation while highest number of cysts was observed after 12 days. Excyctment was observed after 48 hours while encystment started after 8 days. Maximum growth of FLA was observed at 27±2°C. However, both Acanthamoeba and Hartmannella were viable at 50°C for 30 minutes. Trophocidal and cysticidal temperature for Acanthamoeba and Hartmannella was 70°C for 1 min. Disinfection treatment of FLA showed that cysts of Acanthamoeba are more resistant than Hartmannella. The minimum cysticidal concentration (MCC) of chlorine for Acanthamoeba, Hartmannella, and Allovhalkampfia was 2 0 ml/L and 10ml/L, and 4mg/L, respectively, with an exposure time of 30 min. MCC of H<sub>2</sub>O<sub>2</sub> was 7.5% and 10% for Acanthamoeba and Hartmannella, respectively. Comparison of disinfection treatment showed that chlorine is more efficient against cysts of FLA. The present pioneer study in Pakistan will help in water treatment to target FLA.

#### ECONOMICAL PRODUCTION OF CITRIC ACID FROM VEGETABLE WASTE

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Pertaining to essential industry wide usage citric acid is produced through several methods. This paper led to investigate the production of citric acid by using dehydrated and non-dehydrated vegetative waste as a substrate by *Aspergillus niger* through submerged fermentation. During several stages of the production, it was witnessed that maximum production of citric acid was attained by the sweat potato with glucose 50g/L and potato with fructose 42.24g/L in fermented broth at 30 °C for 11 days. The partial recovery of the citric acid was accomplished by the crystallization and estimated by High Performance Liquid Chromatography.

# SESAME OIL MITIGATES ASPARTAME CAUSED OXIDATIVE STRESS, BIOCHEMICAL CHANGES AND HISTOPATHOLOGICAL LESIONS IN KIDNEYS AND LIVER OF ALBINO MICE

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Aspartame is an artificial sweetener used by more than two hundred million people worldwide and is one of the most disputatious food additive, having contradictory evidences regarding its safety. Ongoing research will highlight possible cytotoxic effects of aspartame and protective potential of sesame oil in rodent model. Forty male albino mice weighing 26±2g were randomly and equally categorized into four groups. Mice were exposed to 40μg/g/day of aspartame with and without sesame oil in various groups. Doses were administered through oral gavage for 60 days routinely. At the end of experiment dissections were carried out to collect blood samples for serum biochemistry, liver and kidney tissues for histopathology and to evaluate some anti-oxidative and oxidative stress markers from liver and kidney homogenates. There was no mortality or moribund mice recorded during the whole experiment. Mice body as well as organ weight increased in aspartame group as compared to control. Elevated level of AST, ALT, ALP, Urea and Creatinine were significantly observed in blood plasma of aspartame group against control. Histopathological defects showed ballooning hepatocytes, vacuolations, congested central vein and proliferation of kuffer cells in liver tissues and renal hemorrhage, increased capsular space and glomerulosclerosis in kidneys of aspartame treated mice. Aspartame exposure resulted as oxidative stress by decreasing GSH, SOD, catalase and increasing MDA levels in kidney and liver tissues. Besides, co-administration of sesame oil with aspartame led to significant protection against aspartame induced toxicities. Hence aspartame can instigate biochemical and histopathological alterations in albino mice, while sesame oil has potential to forfend these.

# STUDY ON MORPHOMETRIC FEATURES OF DIFFERENT REGIONS OF EPIDIDYMIS IN DAMANI GOAT BUCK

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Epididymis play vital role in reproduction of livestock. In order to documents the exploration of the potential reproductive capabilities of the local animal resources, the current study was carried out on dammani bucks in the subtropical environment of Peshawar. The current research comprised of the detail exploration of the three segment of epididymis in relation with appraisal the gross morphometry along with the histological characteristics of each segment. Local slaughter houses in Peshawar was used for collection of the specimen. Standard protocol was adopted in the collection and transportation of the specimen to the laboratory of vet. Anatomy, CVS. The gross morphteric examinations included length, width, weight of each segment were carried out through standard procedure via Vernier caliper and measuring tape and electric weighing machine respectively. Likewise, for histological investigation of the three segment of the epididymis, routine standard histopathological procedure were used. Furthermore, Protein expression in the three segment of the epididymis were carried out in the proteomic section in Histopathology laboratory using recently developed protocol. During current, the gross morphometric investigation demonstrated the mean length of head of right and left epididymis was 6.72±0.20, 6.26±0.20 cm likewise the length of the body of right and left epididymis was 6.18±0.14, 6.41±0.14 cm and that tail of the right and left epididymis 1.76±0.09, 1.56±0.10 cm of right The histological assessment of this vital organ demonstrated that epididymal duct is lined by epithelium and the duct contains tubular regions wherein diverse biological cells having important reproductive role are abundantly expressed such as the principal cells, narrow cells, basal cells and apical cells.

#### GC-MS ANALYSIS OF LIPID EXTRACT IN MUSEUM BONE OF BLUE WHALE

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Pakistan has a diversity of cetacean population in its offshore seas; these animals comprises of enormous quantity of lipids in their body especially below the epidermis of skin, intact in bones and in liver. Lipids are naturally occurring biological compounds, which are hydrophobic in nature. Museum skeleton of blue whale present in CEMB, were examined to identify the fats found within the bones. Maceration technique was adapted for extraction of lipids. Fatty acid composition of lipids was analyzed through saponification followed by methylation of the fatty acids. Gas chromatography Mass spectrum (GC-MS) was employed to investigate the extract. Digital mass library NIST and MS search 2.0 was used for the matching of spectra. MUFAs, PUFAs and some pesticides have been identified along with major concentration of free fatty acids. The main fatty acids are, C13:0, C15:0, C16:0, C16:1, C14:0, C18:0, C18:1, C20:1 and permethrin. These extracts possess essential fatty acids were also govern numerous biological activities i.e. anti-oxidant activity.

# 2. CELL AND MOLECULAR BIOLOGY, CELL BIOLOGY, GENETICS

#### MOLECULAR BASIS OF HEAT STRESS ON GROWTH OF PARAMECIUM SPECIES

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The preferential induction of Hsp90 in response to heat shock has been shown to vary in *Paramecium* species. Hsps is a kind of heat shock response in organisms to occupy variable thermal niches. The present study reveals the role of Hsp90 as a molecular diagnostic tool to assess environmental stress, thermal tolerance limits and survival of three species of *Paramecium* (*P. multimicronucleatum*, *P. jenningsi*, and *P. primaurelia*). All these species were exposed to constant (34°C for two weeks) and temporally variable temperatures (30°C, 34°C and 38°C overnight). *Paramecium* species have shown their resistance towards the fluctuating temperatures, which was well explained with the help of the present study by showing the presence of heat shock proteins through SDS-PAGE. At 30°C, all species showed expression except *P. jenningsi*. At 38°C, only *P. primaurelia* did not show expression of Hsp90. This suggests that *P. jenningsi* has a higher thermal tolerance limit, great survival, and more resistant species than the *P. primaurelia*. *P. multimicronucleatum* showed expression at all temperatures, which represent a wider thermal range. *P. multimicronucleatum* and *P. jeningsi* showed increased expression of Hsp90 when exposed to heat stress at 34°C for two weeks, unlikely to *P. primaurelia*. This decreased expression of Hsp90 at higher temperatures is due to the increased sensitivity of *Paramecium* species to heat shock and vice versa. These cultured *Paramecium* species can be used in large scale experiments in lab as well as in stagnant wastewater ponds that will be a novel approach in bioremediation.

#### GROWTH PATTERNS OF CILIATES IN THE PRESENCE AND ABSENCE OF AZO DYES

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The release of azo dyes through textile effluents serves as the primary source of water pollution. Therefore, it has become necessary to degrade dyes using microorganisms. Ciliates can effectively employ industrial effluents in bioremediation because they can survive longer in contaminated water, indicating their ability to detoxify pollutants and improve effluent quality. The present study was conducted to examine the effect of an azo dye, Brilliant Yellow, on the growth patterns of *Paramecium caudatum* named AS, at different pH and temperatures and to evaluate the decolorization potential ciliate. *P. caudatum* was cultured in Bold Basal Salt Medium and exposed to varying pH (6, 6.5, 7, 7.5, 8) and temperature (20°C, 25°C, 30°C, 35°C), respectively. The growth pattern was examined daily for 30 days. All experiments were performed in triplicates. AS dye-treated samples showed maximum growth of 6400 cells/ml at pH 7 and AS control samples showed the best growth of 8400 cells/ml at pH 7.5, whereas AS control and dye-treated samples each showed maximum growth of 5400 cells/ml at 25°C. The results indicated that *P. caudatum* showed 86.66% decolorization. Products formed after dye degradation was analyzed by Fourier-Transform Infrared Spectroscopy (FTIR), which confirmed the biotransformation of this dye. Therefore, ciliates can be used for the decolorization of industrial wastewater containing azo dyes.

### TOXIC EFFECT OF AZO DYES ON THE ACTIVITY OF VARIOUS ENZYMES AND METABOLITES IN CILIATES

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The contamination of water bodies by numerous anthropogenic activities has highlighted the need to identify new bioassays to assess water pollution. The freshwater ciliate *Paramecium* has been shown to play a key role in the biodegradation of dye-containing wastewater. The present study was conducted to evaluate the toxic effects of an azo dye (Brilliant Yellow) on the activity of various antioxidant enzymes and other metabolites in two species of ciliates *i.e. Paramecium multimicronucleatum* (IZ) and *Paramecium caudatum* (AS). Both the species were cultured in bold basal salt medium, and the treated species were exposed to the Brilliant Yellow azo dye when optimum growth for each species was attained. Total protein content in both IZ and AS treated species was considerably increased compared to their control. Elevated levels of GSH were reported in both treated species of IZ and AS. After treatment with BY dye, GST activity was also increased in both species, but it was greater in IZ (50%) than AS (33.33%). Likewise, catalase activity was also relatively higher in IZ (137.76%) than in AS species (91.48%). In contrast, superoxide dismutase and glutathione peroxidase activity were recorded to be higher for AS treated species (56.52% and 36.76%) than IZ treated species (35.71% and 22.22%), respectively. Thus, a strong induction in the activity of antioxidant enzymes after exposure to azo dye suggests that ciliates can be extensively employed as potential bioremediators for treating dye-containing wastewater.

# EFFECT OF AZO DYES ON THE GROWTH PATTERN OF CILIATES BY CHANGING VARIOUS PH VALUES

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Environmental health has been greatly affected in recent decades by the discharge of azo dyes and their metabolized products from various industries. Textile effluents containing azo dye can be treated by bioremediation, offering many advantages over standard treatment methods. Ciliated protozoans have been used as bioremediation tools due to their easy maintenance under laboratory conditions, faster growth rates, and lack of cell walls. The current study investigated the growth phase, the effect of azo dyes on growth of *Paramecium jeninjsi* at varying pH (6, 6.5, 7, 7.5, 8 and 8.5) and temperature (20, 25, 30 and 35°C) and its decolorization potential. *Paramecium* samples were provided by the Molecular Biology Lab at the Institute of Zoology, University of the Punjab, Lahore. For studies of growth phases, *Paramecium* culture was maintained at a pH of 7.2 and a temperature of 25°C. Growth was monitored on daily basis for 15 days. The growth of *P. jeninjsi* were observed at different pH and temperature of treated and control samples for 30 days. The maximum growth of *P. jeninjsi* was observed at pH 7 and temperature 25°C *i.e.*, 2600 and 3200 cells/ml in control and treated samples, respectively. 85% decolorization was observed in treated samples after 6 days. Analysis of dye degradation products was carried out using FTIR. The experiments were performed in triplicates. Results revealed that *P. jeninjsi* can be used as efficient bioremediation tool in cleaning wastewater containing toxic azo dyes.

# A STUDY OF LEVEL OF FASTING BLOOD SUGAR, SERUM INSULIN AND INSULIN SENSITIVITY INDEX IN OBESE POSTMENOPAUSAL WOMEN OF KARACHI, PAKISTAN

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The prevalence of obesity in women around and after menopause is high and is rising worldwide, changes in hormone levels can trigger fluctuations in blood sugar level and insulin resistance. This study was planned to examine the differences in BMI, FBG and insulin level in obese subjects as compared to normal weight. This experiment was carried out in Karachi city of Pakistan between, January 2015 to April 2015. We included women aged 45-60 years (n=84) with ceased menstruation for at least 12 month and had a natural menopause, Anthropometry measurements were taken to calculate BMI and WHR. A fasting venous blood sample of 5ml was taken and serum was separated and stored. FBG level was measured by using GOD PAP method, while Serum insulin, was measured with ELISA. Insulin index was calculated by formula using QUICKI method. Unpaired t-test was used to compare the mean of control (n=13) and obese (n=71) group. Our findings evaluated the significant differences in the average BMI values of control (22.14  $\pm$  0.023) kg/m<sup>2</sup> and obese postmenopausal women (35.93  $\pm$ 0.72) kg/m<sup>2</sup> BMI of obese females were significantly higher than control females (P<0.001). Mean FBG level of control (92.15  $\pm$  1.48) and obese postmenopausal women (117.56  $\pm$  0.95) mg/dl were significantly higher in obese group (P<0.001). Serum insulin level and insulin sensitivity index of control females were averaged at  $24.13 \pm 3.03$ and  $0.30 \pm 0.006$  respectively. While serum insulin mean values and insulin sensitivity index of obese women were  $40.37 \pm 5.07$  and  $0.28 \pm 0.003$  respectively. Both the parameters were related significantly in two groups (P<0.05) and P<0.05) respectively. This study concluded that FBG, serum insulin and its sensitivity index are elevated in obese women. Menopause and adiposity are more common factors that put the women at a risk of developing diabetes. Lifestyle modifications play a major role in controlling obesity and fluctuated FBG and insulin levels.

### ASSOCIATION OF DIABETES AND SERUM VITAMIN D LEVELS IN OBESE/ OVERWEIGHT DIABETIC AND NON-DIABETIC WOMEN

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The world has been facing a pandemic namely "Type 2 Diabetes Mellitus" (T2DM) since ages with extremely high prevalence in almost every population. It is more commonly found among obese and overweight individuals with females being a constant target. The purpose of this study is to investigate three variable based hypothesis patterns – Vitamin D deficiency induces obesity developing into T2DM, along with the consideration of physical and clinical parameters in Pakistani women Lahore. The study group included 60 diabetic whereas the control group consisted of 60 non- diabetic healthy women. Subjects of both groups were obese/ overweight with BMI more than  $25 \text{kg/m}^2$  and of similar age ranging from 30-70 years specifically with vitamin D deficiency. Physical parameters including BMI, diet, rate of physical activity, sunlight exposure and family history were obtained through questionnaires. The clinical factors including serum vitamin D, calcium and fasting glucose were obtained through blood serum and whole blood testing, respectively. Results showed that 76% of healthy women had glucose levels more than 100 mg/dL indicating a high risk of diabetes onset, whereas the experimental group already had high levels with the mean range of 223 mg/dL and Std. deviation  $\pm$  50.2. Non-diabetic females had more Vitamin D3 deficiency with mean value 20 ng/ml with Std. deviation  $\pm$  19.7 compared to diabetic group with mean insufficiency value of

25.5 ng/ml and Std. deviation  $\pm$  10.8. Both groups were observed to have majority women with normal calcium levels with mean values ranging from 9.2 to 9.3 mg/dL. The obtained clinical values mark an abnormally reversible metabolism. Majority of the healthy females were found to be at risk of diabetes development due to an extremely lower rate of vit D accompanied by similarities with the study group in various parameters. Results concluded that there is a weak positive significant difference between Vitamin D, calcium, BMI of both diabetic and nondiabetic overweight patients.

#### STUDY OF HEAT SHOCK PROTEINS (HSP20) IN TROGODERMA GRANARIUM

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Trogoderma granarium (Everts), control has become difficult due to the emergence of resistance against almost all the known insecticides and fumigants. Moreover, they have hazardous effects on human health. Heat treatment has been used to control the khapra beetle infestation in storage goods. However, khapra beetle shows a great tolerance to heat stress. Larval stages of khapra beetles are the most tolerant life stages to thermal stress. In present study, 4th and 5th larval instar of Lodhran and Lahore populations were exposed for 1 hour with increasing temperatures (40°C, 45°C, 50°C, 55°C, 60°C). Larvae exposed to 35°C for 1 hour were treated as control. Characterization of insect homogenate showed that the heat shock protein was one of the major elements in the ability of thermotolerance. By using 10% SDS PAGE analysis, heat shock protein with molecular weight of 25kDa was observed in 4th and 5th larvae of both populations. Results of present study showed that the small heat shock proteins (shsp25) were significantly higher expressed during active larval stages of both populations. The expression of sHsp25 was significantly upregulated with increasing temperatures (40°C, 45°C, 50°C) but its expression diminishes at extreme temperature (60°C). Intensity of bands showed that the Lahore population is more resistant to thermal stress as compared to Lodhran population. The expression of heat shock protein in response to increasing temperature appears to be associated with the ability of khapra beetles to survive in response to heat shock stress. The current study reported small heat shock protein (sHSP25) in Trogoderma granarium which may give insights to the researchers for the control strategies of this invasive pest.

## DISCOVERY OF DIFFERENTIALLY EXPRESSED GENES OF *PARAMECIUM* SP. UNDER COPPER IONS STRESS USING TRANSCRIPTOME SEQUENCING

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Ciliates are mostly present in the heavy meatal contaminated environments and have developed mechanisms to survive in it. Due to this ability, they secure prominent position among organisms used for bioremediation of heavy metals. Nowadays, *Paramecium* is largely used for the bioremediation studies. To investigate the response of *Paramecium* at gene level, transcriptome sequencing was done under copper exposure. RNA sequences from *P. multimicronucleatum* were assembled with Trinity program. Transcript abundance and differential expression of genes was analyzed by RSEM and edgeR. Taxonomy of the data was verified using Blastx and diamond with MEGAN. Contaminated sequences base on taxonomy removed from the data. Genomic data revealed 18, 401 sequences from *Paramecium multimicronucleatum*. Copper up regulated genes are 1,172 identified with significant value 0.5 and log<sub>2</sub> Fold change. Annotations obtained from three protein databases UniProt, Pfam and InterPro. Most of the genes were trichocyst matrix proteins and 70% identical to amino acid sequences from *P. tetrraurelia*. Other stress dealing genes including Glutathione S transferase, thioredoxins and heat shoch proteins were upregulated under copper exposure as compared to control. This shows that stress related genes and pathways are upregulated to deal with the heavy metal for the survival of cell.

# ASSOCIATION OF *IRGM* PROMOTER REGION POLYMORPHISMS AND HAPLOTYPE WITH PULMONARY TUBERCULOSIS IN PAKISTANI (PUNJAB) POPULATION

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The human immunity related GTPase M (IRGM) is a key factor in autophagy. Single nucleotide polymorphisms (SNPs) in IRGM have been reported to affect the Mycobacterium tuberculosis (M.tb) degradation pathway, causing M.tb cellular retention and promoting tuberculosis (TB) pathogenesis. The objective of our case-control study was to screen the promoter region of IRGM gene for regulatory SNPs, in a Pakistani population including 70 patients and 30 healthy controls (15 control TB non-exposed (Ctrl-TBne)) and 15 control TB-exposed (Ctrl-TBe)). DNA was extracted from blood, and the upstream promoter-enhancer amplified and sequenced. Our findings showed group specific variations in allelic and genotypic frequencies at four loci: -1161T/C (rs4958843), -1133G/A (rs4958423), -1049C/T (rs4958424) and -708G/A (rs35707106). Allele G (p-value = 0.027) and genotype GG (p-value = 0.04) at -1133G/A; allele C (p-value = 0.029) and genotype CC (p-value = 0.05) at -1049C/T; and allele G (p-value = 0.02) and genotype GG (p-value = 0.04), showed significantly higher association with TB patients as compared to healthy controls (both Ctrl-TBe and Ctrl-TBne). At -1161T/C, allele T was found to be more frequent in patients (p-value = 0.03), but no difference in genotypic frequency was found among the groups. These SNPs display strong linkage disequilibrium (LD). Haplotype analysis of these SNPs yielded ten haplotypes, of which -1161T/ -1133G/ -1049C/ -708G (p-value= 0.007) was found to be associated with TB status. This 4-SNP haplotype also represents an Expression Quantitative Trait Locus (eQTL), associated with Crohn's disease and chronic inflammatory diseases. Our findings support the hypothesis that variants -1161T/C, -1133G/A, -1049C/T, and -708G/A are associated with IRGM expression and susceptibility to TB in a Pakistani (Punjab) population.

#### PREVALENCE OF VARIOUS GENES CAUSING EPILEPSY IN PAKISTANI POPULATION

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Epilepsy is the neurological disorder that indicates abnormal activity in central nervous system. It is caused due to genetic as well as non-genetic factor. Prevalence rate is higher in non-developed countries. In Pakistan the prevalence of epilepsy is about 9.99/1000. Different risk factors such as head trauma, central nervous system infections, poverty and tumors are associated with development of epilepsy in different age groups. This study was performed to identify pathogenic variants by WES in Pakistani Population. In the present study patients with epilepsy were sort out from different hospitals of Punjab, Pakistan from 1st December 2017 to 31st August 2019. We performed next generation sequencing with epilepsy patients. We identified three missense pathogenic or likely pathogenic mutation in three genes such as heterozygous c.4442A>G (p.Asp1481Gly) mutation in DMXL2 gene, heterozygous c.1991T>C (p.Val664Ala) mutation in CACNA1H and homozygous c.6695A>G (p.Tyr2232Cys) mutation in GPR98 gene. All patients showed generalized tonic colonic seizure. Different parameters show different frequency. All patients have showed different onset age. Frequency of tonic colonic seizure is higher as compared to other seizure types. Frequency of seizure onset age was higher between 11-20 years of age. In present study disease

prevalence is higher in male as compared to females. Patients that used antiepileptic drug showed highest frequency as compared to those that do not use medicines. These all are already reported genes which involved in epilepsy. All parameters showed association with reported literature.

# EFFECT OF INHIBITOR UNC0642 ON EXPRESSION OF G9A AND ITS PROLIFERATIVE AND APOPTOTIC MARKERS IN BREAST CANCER CELL LINE MCF-7

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G9a is a lysine methyltransferase that has been associated with various types of cancers. In cancerous cells, the expression of G9a has been reported to be upregulated. Moreover, G9a has been reported to downregulate expression of tumor suppressor genes in a variety of cancerous cells. Therefore, making it a target for anti-cancerous therapies. While various inhibitors of G9a are under study, the present study explores the anti-cancer activity of UNC0642, a novel potent inhibitor of G9a. UNC0642 treatment at various concentrations i.e., 1μM, 5μM, 10μM, 15μM, and 20μM was given to MCF-7 breast cancer cell line. Analysis of growth and proliferation parameters by various cellbased assays such as neutral red assay, MTT assay and BrdU cell proliferation assay showed that lower concentrations of UNC0642 were less cytotoxic to the cells whereas the higher concentrations were lethal for the cancerous cells. IC50 was found to be 12.6µM. Further contextualization of therapeutic potential of G9a inhibition was provided by expression analysis of G9a gene and genes associated with cell proliferation, tumor suppression, and apoptosis. The gene expression of fibroblast growth factor 1 (FGF1), enolase 2 (ENO2) cyclin D1 (CCND1), catalytic subunit of AMP-activated protein kinase (AMPKα2), RNA polymerase II transcription elongation factor (ELL2), and pro-apoptotic Bcl-2 homology 3-only protein (BIM) in MCF-7 breast cancer cell line and normal human embryonic kidney HEK-293 cells was studied in response to treatment with the aforementioned concentrations of UNC0642. Inhibition of G9a expression was observed in dose-dependent manner in both cell lines with increasing UNC0642 concentrations. A dose-dependent downregulation of gene expression of proliferation marker genes (FGF-1, CCND1, and ENO2) was observed in both cell lines, whereas upregulation of tumor suppressor genes (AMPKα2 and ELL2) and apoptotic marker gene (BIM) was observed in dose-dependent manner. The results indicated the potential of G9a inhibition in reduction of cancerous cell proliferation and inducing apoptosis in cancerous cells. Further elucidation of the signaling pathways associated with G9a, in-vitro and in-vivo safety analyses is required to fully establish G9a as a therapeutic target for cancer progression and UNC0642 as an anti-cancerous therapeutic agent for treatment of breast cancer.

# EFFECT OF METFORMIN ON P53 AND ITS INTERACTING PARTNERS IN BREAST CANCER CELL LINE MCF-7

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Metformin is known to favor p53 anti-cancer activity and promotes apoptosis and cell senescence in the case of cancer cells. It decreases the incidence of several cancers but the findings regarding breast cancer are mixed. In the present study, the effect of metformin on the expression of p53 and its interacting partners MDM-2, PIRH2  $\Delta$ Np73 was studied on breast cancer cell line MCF-7 and it was compared with normal kidney cell line HEK293 after providing metformin treatment of different concentrations to both cell lines. Analysis of the cytotoxicity and proliferation parameters showed that higher concentrations of metformin were cytotoxic for the breast cancer cells with EC<sub>50</sub> of 22.75 $\mu$ M. Analysis of expression of p53 and the interacting genes showed that metformin upregulate p53 expression in dose-dependent manner, while the expression of interacting partners is downregulated indicating that metformin exhibits the anti-cancerous properties by modulating p53 pathway. Altogether this study for the first

time reported the expression analysis of p53 and its interacting partners in breast cancer cell line MCF-7 after metformin treatment that can help provide protective effect of metformin on breast cancer risk, however further investigation is needed to ensure the link between decreased breast cancer risk and metformin.

# HEAT SHOCK PROTEINS ARE EXPRESSED IN HIGHER QUANTITIES IN LONGEVITY INDIVIDUALS OF D.I. KHAN DISTRICT

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The cellular response to stress is mediated by intracellular proteins, called Heat Shock Proteins (HSPs). Among various known stressors, heat is a major factor that induces the production of HSP70. Keeping in view the very hot conditions of Dera Ismail Khan (D. I. Khan) division where the temperature remains at 45-50°C during the months of June to September, it was hypothesized that heat stress conditions do induce the overexpression of HSPs, especially Hsp70. It was thus attempted to find out the possible role of Hsp70 in those human being having the age of 90 years or above, called longevity people, against heat stress conditions. Whole blood samples of 45 longevity individuals and 20 samples of control people were collected in D. I. Khan during September 2018 to October 2019 after proper approval from Gomal University ethical review board and written consent of each individual was taken prior to collection of blood sample. For serum collection, blood samples were centrifuged at 1000xg for 15 minutes. Quantitative measurement of Hsp70 protein was done using sandwich ELISA technique. The maximum serum HSP70 level observed was 42ng/ml and minimum serum Hsp70 value was 13ng/ml, with median value of 28ng/ml. In control group, maximum concentration observed was 38ng/ml while the minimum serum level was 18ng/ml with median value of 13ng/ml. In longevity males, serum Hsp70 levels increased in individuals between 89 to 91 years of age, peaked between 92 to 97 years but comparatively lower having the age of above 98 years. On the other hand, serum concentration of Hsp70 in longevity females were highest in those having 89 to 92 years, lower in 93 to 97 years and, like males, lowest having age above 98 years. The study hence, showed that longevity individuals had higher quantities of serum Hsp70 compared to control group, and again, males contained higher concentration than longevity females, showing that thermal stress was the agent leading to over-expression of Hsp70 especially in longevity individuals. Whether this increased expression had any impact on longevity is still not clear that needs to be deciphered.

# MOLECULAR IDENTIFICATIONS OF FAMILY RANIDAE FROM CHANGA MANGA FOREST, PUNJAB, PAKISTAN

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Identification of amphibians on morphological basis is still considered authentic. However, molecular analysis of these taxa is required for exact species identification. During present survey 48 amphibian specimens were captured from the study area. The captured specimens were represented by 2 species, 2 genera belongs to 1 family. The average SVL of *Hoplobatrachus tigerinus* and *Euphylctis cyanophylictis* was 82±22.13 mm and 44.25±2.197 mm, SL was 5.7±1.18 and 3.15±0.455, HL was 27.85±5.92 and 13.05±0.761, ED was 4.55±0.74 and 3.6±0.458, HTYD was 6.67±1.57 and 3.34±0.475, VTYD was 6.22±2.04 and 3.4±0.490, HAL was 15.355±1.62 and 12.58±0.381, FTL was 37.55±6.54 and 22.69±0.909 and average body weight was 88.11±70.53 and 14.57±3.370. Total genomic DNA of *Hoplobatrachus tigerinus* and *Euphylictis cyanophylctis* was extracted from blood. Phylogenetic trees were constructed viz., the neighbor-joining tree, the maximum likelihood tree and parsimony using MEGA 10 to check relationships among species. The obtained DNA sequence has shown reliable and clear species identification. After trimming ambiguous bases, the obtained 16S rRNA fragment of *Hoplobatrachus tigerinus* and *Euphylctis cyanophlyctis* was 536 and 515 bp while 16S rNRA fragments aligned with NCBI sequences

comprised 510 bp. The intraspecific variation of *Hoplobatrachus tigerinus* is 3-7% and the intraspecific variation of *Euphlyctis cyanophylctis* is 17%. The interspecific variations between *Hoplobatrachus tigerinus* and *Euphlyctis cyanophylctis* is 7-17%.

## MOLECULAR PHYLOGENETICS OF FAMILY ELAPIDAE FROM DISTRICT KASUR, PUNJAB, PAKISTAN

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Molecular based identification and phylogenetic relationships of family Elapidae is still unclear in Pakistan. Present study is therefore planned to explore and identify species of family Elapidae from district Kasur, Punjab. Specimens of Common krait (*Bungarus caeruleus*) and Cobra (*Naja naja*) were collected from selected sites of study area and each captured specimen was tagged with specific number. Three specimens of each species were euthanized and preserved in 75% ethanol for molecular characterization. Total genomic DNA was extracted from preserved tissues by using phenol chloroform method. Purity of DNA was checked through agarose gel electrophoresis. Extracted DNA was amplified using 16S rRNA primer set. The obtained DNA sequences have shown reliable and clear species identification of all the captured specimens. After trimming ambiguous bases, the obtained 16SrRNA fragment of *Bungarus caeruleus* and *Naja naja* was 690 bp and 666 bp respectively. Recently few DNA barcoding studies of Asian reptiles have been carried out and sequences for related species were available at NCBI. Closely matched DNA sequences were retrieved from NCBI in blast searches and incorporated in N-J tree. The overall, genetic divergence of common krait and cobra was 0.155±0.044 and 0.023±0.013 respectively.

# EFFECT OF BUTYRATE ON GENE EXPRESSION OF AKT AND MTOR-INVOLVED IN REGULATION OF CELLULAR PROLIFERATION IN MURINE BRAIN

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Butyrate is a short chain fatty acid produced naturally by gut microbiota. It has a significant potential to inhibit the expression of histone deacetylase complex thus controlling the regulation of many genes. Butyrate exerts a beneficial neuropharmacological effect on brain disorders resulting in reduced risk of neurodegenerative diseases. Akt/mTOR axis is important in maintaining cellular growth and proliferation where changes in nutrient availability would lead to fluctuation in metabolic energy homeostasis. The current study compares the expressions of Akt and mTOR in murine brain tissues having orally and intraperitoneally administered NaB (butyrate salt) and metformin with those of control group. 15-18 weeks old mice were given drugs dosage after every 24 h for 16 days. The mice were sacrificed and internal organs were dissected out. Total RNA was extracted from brain tissues and used for cDNA synthesis. Real time PCR was performed to measure relative gene expression. Results showed the increased expressions of Akt (fold=1.93; p=0.04) and mTOR (fold=1.84; p=0.07) in response to oral NaB treatment. The intraperitoneal NaB resulted in slightly decreased expressions of Akt (fold=0.81; p=0.8) and mTOR (fold=0.95; p=0.8). In metformin group, expressions of Akt (fold=0.71; p=0.2) and mTOR (fold=0.70; p=0.4) were decreased in oral treatment. Similarly, expressions of Akt (fold=0.88; p=0.7) and mTOR (fold=0.50; p=0.09) were decreased in intraperitoneal administration. This study revealed differences in the pattern of genetic expression with oral and intraperitoneal route of NaB in mice. Whereas metformin showed decreased expressions of Akt/mTOR axis in both routes of drug dosage. Increased expressions of Akt/mTOR axis in oral NaB can be related to provide more energy and survival signals for the growth and proliferation of healthy cells. Decreased expressions of Akt/mTOR axis in mouse models have anti-cancer effect in regulating the apoptosis of abnormal cells to prevent their further growth and proliferation.

# EFFECT OF BUTYRATE NN GENE EXPRESSION OF SIRT1 AND AMPK - INVOLVED IN NUTRIENT SENSING AND ENERGY HOMEOSTASIS - IN MURINE BRAIN

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Butyrate has been characterized as a histone deacetylase inhibitor. It causes acetylation of histone altering expression of regulatory proteins and several transcription factors, thereby affecting cellular energy homeostasis and proliferation. Butyrate restores brain functions and exhibits several neuroprotective effects in many neurodegenerative diseases. Sirt1 and AMPK can reciprocally enhance each other's activity and have a crosstalk in regulation of metabolic responses. In current study, murine brain cells were used to determine the effect of NaB (butyrate salt) on gene expression of AMPK and Sirt1. Mice (15-18 weeks old) were treated with NaB (experimental group), metformin (positive control) and PBS (negative control); all given orally as well as intraperitoneally (I.P) for 16 days. Total RNA was isolated followed by cDNA synthesis and gene expression was determined by real time PCR. The expression of AMPK was increased 1.62 (p=0.3) and 2.81 folds (p=0.01) in response to oral NaB and metformin, respectively. Sirt1 expression remained unchanged by NaB but was increased by 2.12 folds (p=0.1) in response to oral metformin treatment. Similarly, in case of I.P treatment, NaB and metformin groups showed insignificantly decreased genetic expression of AMPK; 0.78 and 0.90 folds, respectively. The expression of Sirt1 was increased by 2.88 folds (p=0.3) and 1.22 folds (p=0.3) in response to I.P NaB and metformin treatments, respectively. These results showed that NaB exerts same effect on AMPK/Sirt1 axis in murine brain as that of metformin. This study constitutes a step forward in understanding the neuroprotective role of NaB through enhanced expression of factors involved in AMPK/Sirt1 axis.

### CLONING AND CHARACTERIZATION OF CRY11 CRYSTAL PROTEIN FROM BACILLUS THURINGIENSIS

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Bacillus thuringiensis is an entopathogenic soil dwelling bacteria which produce cry and cyt toxin and are being used as a bio pesticide rather than chemical insecticide to control resistance in insects. The present study describes the screening of toxic Bt. (9NF, 4NF, 6NF) isolates having 99% homology with Bti. prototoxin Bacillus thuringiensis (AXJ97553.1 & novel OUB27301.1) which harbouring full length cry11 gene (1.9kb). Full length cry11 (1.9kb) gene was initially cloned in pTZ57R/T and subcloned in pET-30a (+) for expression. The suitable optimized condition IPTG induction was 1mM and incubation period was ranging 3.5 -4 hours at 37°C to get good expression. Bioassays were performed with expressed purified and crude recombinant protein against 3rd instar larvae of Aedes aeygpti using recombinant organism (E.coli BL21, DE3) transmuted with cry11 gene. Purified Bt. protein is most toxic with LC50 value of 42.883±6 μg/ml against dipterans larvae.

#### PREVALANCE OF HEPATITUS B AND C IN THALASEMIC PATIENTS OF BAGH

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Thalassemia is a well-known inherited hematologic disorder caused by a decrease or an absence of globin production. Thalassemia patients depend upon regular blood transfusion for maintaining their hemoglobin level. Due

to regular blood transfusion in these patients their lives saves, but increases the risk of getting viral infections like hepatitis B virus, hepatitis C virus and human immunodeficiency virus. The aim of this study was to find out the relative abundance of HBV and HCV in some districts of Azad Jammu and Kashmir. During the study period, a total of 145 Thalassemic patients were enrolled in two different centers (hospitals) of Azad Jammu and Kashmir. All of these patients were previously admitted and regularly receiving blood in respective centers. Out of them, 81 were registered in Sheikh Khalifa Bin Zaid Hospital, Rawalakot and 64 in District Headquarter Hospital Bagh, Azad Jammu and Kashmir. Out of the total 145 Thalassemic patients, 15 (10.34%) were positive for HCV while all others were negative and only 3 (3.70%) out of 81 patients were HBsAg positive in Sheikh Khalifa Bin Zaid Hospital Rawalakot while no one was HBsAg positive in District Headquarter Hospital Bagh. The highest number of HCV and HBV positive patients was found in Rawalakot as compare to Bagh. More blood donor screening programs and effective screening techniques are needed to prevent transmission of HCV infection among Thalassemic patients. All Thalassemic patients should be vaccinated for HBV before transfusion. Blood donor screening programs and effective screening techniques are needed to prevent transmission of HBV and HCV infection in Thalassemic patients.

### SEQUENCE OF C-PEPTIDE AND RESIDUES ON THE ACYL-SIDE OF SCISSILE BOND DETERMINES THE CLEAVAGE SPECIFICITY OF TRYPSIN FOR CONVERSION OF PROINSULIN TO INSULIN

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In vivo, proinsulin is processed to insulin using two prohormone convertases, PC1 and PC2and a carboxypeptidase E. The convertasesshow specificity for the cleavage of dibasic (RR and KR)–X bond at A/C and B/C junction. In vitro, these two classes of enzymes are substituted by trypsin and carboxypeptidase B respectively. A kinetic study of trypsin proteolysis of M–proinsulin in the absence and presence of carboxypeptidase B was performed. The proteolysis of M–proinsulin with trypsin alone, initially produced M–insulin–RR and M–insulin–R which preferentially gave Des–30–M–insulin as a major product. But in the presence of carboxypeptidase B along with trypsin, these extra arginine residues, from M–insulin–RR and M–insulin–R, were cleaved immediately by carboxypeptidase B and converted into M–insulin, with minor traces of Des–30–M–insulin. The results suggest the protection of the K<sub>29</sub>–A<sub>30</sub> bond of insulin by the attack of trypsin. This protection could be due to the fact that insulin contains compact secondary structure that does not allow trypsin to reach the K<sub>29</sub>–A<sub>30</sub> bond. However, the unexpected feature in our work is that the mere presence of arginine residues at the C-terminal of insulin B-chain, in M–insulin–RR or M–insulin–R, makes this region less compact and the K<sub>29</sub>–A<sub>30</sub> bond becomes accessible to trypsin.

# CO-EXPRESSION OF CHAPERONIN GROEL TO ENHANCE SOLUBLE EXPRESSION OF RECOMBINANT PROCHYMOSIN IN ESCHERICHIA COLI

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Prochymosin is an inactive form of chymosin that is widely used in cheese preparation due to its higher ability for milk clotting and less proteolytic activity. Traditionally, chymosin has been isolated from the calf's stomach but in modern era as the consumption of cheese increasing worldwide, therefore recombinant DNA technology is useful to produce chymosin at large scale. Previously, it was studied that prochymosin is expressed as inclusion bodies in *E. coli* host system using pET21a vector. The objective of this study was to co-express prochymosin and GroEL proteins in *E.coli* (Rosetta gami) host system under optimized conditions. For this purpose, cDNA of M-prochymosin was cloned in pETDUET-1 alone (M-prochymosin/ pETDUET-1) and in another pETDUET1 expression vector that already contain GroEL gene in one of the MCS of vector (M-prochymosin/GroEL/pETDUET1). Various conditions

(IPTG concentration, post-induction time and temperature) were optimized for better soluble expression of Metprochymosin in *Rosetta-gami* cells. The soluble expression of M-prochymosin was obtained at 25°C temperature with induction at 1 mM IPTG for 48 hours. The results here indicated that soluble expression yield of Mprochymosin was increased by assisting its expression with chaperone, GroEL. These findings were remarkably comparable with the results when Met-prochymosin was expressed alone without chaperone assistance. It can be presumed that it might be a potential tool to enhance solubility of aggregation-prone proteins.

### STUDY ON THE TOTAL PROTEINS EXPRESSION OF DIFFERENT REGIONS OF EPIDIDYMIS IN DAMANI GOAT BUCK

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The provision of animal origin protein to the ever-raising population in the country demands the exploration of the hidden productive and reproductive potential of the local indigenous animal resources. Accordingly, here we attempted to documents the exploration of the potential reproductive capabilities of the damani bucks, especially appraisal of total protein expression in three different region of the epididymis which were associated with diverse reproductive functionality including semen maturation and preparing for the fertilization capability. Experiments were carried out on the epididymis specimen of the adult dammani bucks that were collected from the local slaughter houses in Peshawar. The specimens were brought the laboratory of Vet. Anatomy and histopathology, CVS. Protein expression in the three segments of the epididymis were carried out in the protemic section in Histopathology laboratory using recently developed protemic protocol. During current proteomic study, structural protein as indicate 70 and 100 were predominantly expressed in all the three regions of the epididymis. The protein associated with secretary activities were also expressed in the three regions of the epididymis as indicated with 35. Additionally, the protein having relevance with both structure and secretion were expressed in the head, body and tail segment of the epididymis in the damani buck as indicated 15 and 25 respectively. Thus the current study demonstrated the significance of the protein expression profile in the different region of the epididymis in the dammani buck that have the relevance in the enhancement of the reproductive management and improvement to explore the hidden potential of the local animal resources.

# MOLECULAR CLONING AND CHARACTERIZATION OF THE GENE CODING FOR THE NADH DEPENDENT AZOREDUCTASE (AZK) FROM *KLEBSIELLA PNEUMONIAE GM04*

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Eco-friendly and cost effective approaches are needed to remediate dye-contaminated wastewater from various industries before their release into the ecosystem. In this study, a local bacterial strain was isolated from a textile wastewater, capable of decolorizing disperse blue 284 dye and identified as *Klebsiella pneumoniae*. For maximum decolorization the conditions was optimized. The DNA was isolated by phenol-chloroform method and azoreductase gene was amplified by using specific primers. The amplified gene was ligated into cloning vector. The gene sequenced was submitted to NCBI and under accession number MT758472. The gene was further cloned in expression vector pET21a and transformed into BL21C<sup>+</sup> for expression. The optimum condition for expression of AzK was obtained at 0.5mM IPTG for 6hr. Three dyes DB-284, RRR and BGL were decolorized by crude extract and purified AzK enzyme analyzed by UV-vis spectrophotometry. The purified enzyme showed maximum decolorization 96% as compare to crude extract 76%. The metabolites were confirmed by GC-MS analysis.

#### IDENTIFICATION OF NOVEL NATURAL STAT3 INHIBITORS BY VIRTUAL SCREENING

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Cancer is the second leading cause of death worldwide, characterized by disruption of various epigenetic, genetic, metabolic and signaling pathways. Signal transducer and activator of transcription 3 (STAT3) is one of the most important oncogenic transcription factor and constitutive activation of STAT3 play an essential role in cancer initiation, proliferation, metastasis, progression, chemo-resistance and immune invasion. Therefore, it has become an attractive therapeutic target for cancer drug development. In this context, 100 compounds from natural compound library were screened out against STAT3 protein. By docking analysis, twelve compounds were found to target STAT3 protein but three compounds including solasodine, euscaphic acid, and cichoriin appeared as top candidates with high binding energies -8.4, -8.4, -7.9 kcal/mol respectively in comparison with well-known commercially available STAT3 inhibitor S3I-201 (-7.6 kcal/mol). These compounds inhibited the constitutive activation of STAT3 by directly binding to DNA binding domain of STAT3 and impaired targeted genes expression. Moreover, pharmacokinetic properties of these compounds by ADMET (Adsorption, distribution, metabolism, excretion, toxicity) analysis also spoke for their efficacy in pharmacodynamic activities these findings collectively suggest that solasodine, euscaphic acid, and cichoriin are novel potent anticancer agent and may be worth for developing anticancer therapies after further investigations.

#### EFFECT OF LEVOFLOXACIN ON COMPLETE BLOOD COUNT PROFILE SHIFA RABBANI

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A total number of 10 young individuals ranging from 20 to 30 years of age were included in this research from the University of the Punjab during the months of December 2019 to February 2020. The subjects were divided into two groups; one was the controlled group in which the blood samples were taken from the subjects without any drug treatment while the other one was the experimental group in which the subjects were treated with 6 doses of Levofloxacin (500mg) while they were suffering from common flu and then their blood samples were recollected after using Levofloxacin for three days. The blood samples from the controlled and experimental groups were analysed for complete blood count profile. The data demonstrated that the level of Haemoglobin (Hb), Mean Corpuscular Haemoglobin Concentration (MCHC) and Neutrophils was lower a bit in experimental group as compared to the controlled group. Moreover, the number of Red Blood Cells (RBCs), Haematocrit value and Eosinophils increased in experimental group as compared to the controlled group. The other parameters; Lymphocytes, Platelets, Mean Corpuscular Volume (MCV), Mean Corpuscular Haemoglobin (MCH) and Monocytes were significantly lower in experimental group as compared to the controlled group but there was almost the same average of White Blood Cells found in both of the groups. To conclude, the findings of present study demonstrated that Levofloxacin could affect complete blood count profile of human.

# CLINICAL CHARACTERISTICS AND FACTORS ASSOCIATED WITH AMENORRHEA IN FEMALE POPULATION IN PESHAWAR

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Amenorrhea is the absence of menstrual bleeding. In females of reproductive age, diagnosing amenorrhea is a

matter of determining etiology behind it. The aim of present study was to investigate factors associated with amenorrhea in female population in Peshawar. A total of 200 patients (aged 15-39) with amenorrhea, who visited the department of Gynecology Health Care Center Peshawar between July, 2020 and March, 2021 were included in this study. Questionnaire is designed including different factors. 2% of patients were suffering with primary amenorrhea and 98% were suffering with secondary amenorrhea. The secondary amenorrhea was higher in married females (85%) than unmarried (14.5%). The common symptoms among patients were stress (78.7%), PCOS (30%), mood swings (81.2%), bloating (81.7%), fatigue (81.2%), Hirsutism (64.0%), acne (51.3%), pelvic pain (56.9%) and osteoporosis (17.3%). Only 18.8% patients perform physical activity while 90.5% don't have habit of exercise. Primary infertility was also higher than secondary infertility among the analyzed patients. Only 2% of patients gave a history of diabetes and 13.7% patients reported for galactorrhea only. The study reported high prevalence of secondary amenorrhea due to PCOS, abnormal hormonal balance, stress and obesity. Since the complications of amenorrhea take considerable time to develop, it is recommended that females should visit to the doctor early to prevent future complications like infertility.

# SEQUENCE ANALYSIS, PHYLOGENETIC TREE ANALYSIS, SECONDARY STRUCTURE PREDICTION AND 3D-HOMOLOGY MOLECULAR MODELING OF *ORYCTOLAGUS CUNICULUS* (RABBIT) LEPTIN

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Leptin is a member of a cytokine family synthesized by adipocytes and primarily functions against obesity. Leptin reaches the central and peripheral nervous systems, and acts by binding with leptin receptor and activating it, thus regulates appetite and food ingestion, discharge of insulin, basal metabolism, bone mass and reproduction. Recent work was conducted to study sequence analysis, multiple sequence alignment and phylogenic tree analysis of leptin. This study predicted the secondary structure of leptin family proteins. The multiple sequence analysis of rabbit leptin showed that out of 167 amino acid residues, 68 positions are found conserved. Phylogenic tree analysis has shown a close relationship between African elephant, woolly hare and rabbit leptin. The predicted 3D structure of rabbit leptin shows the presence of four alpha-helices. At positions 94 and 144, and 59 and 117, functionally important and conserved cysteine and tyrosine residues are present, respectively. Mutation analyses in human leptin has shown the presence of certain amino acids in rabbit as wild type. These finding suggests that rabbit can be used as promising model to study human leptin related diseases.

#### ANTI-ADIPOGENIC POTENTIAL OF CISSUS QUADRANGULARIS

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Obesity is a multifaceted disease which involves an excessive amount of body fat. It is consider as a medical problem that increases the risk of other diseases and health problems, such as heart disease, diabetes, high blood pressure and certain cancers. There are severe side effects of obesity medication approved by FDA. The herbal alternative for weight loss medication will not only provide effective reduction in BMI (body mass index) but also

will be free of severe side effects. Cissus quadrangularis (CQ) has potential to be used as alternative to other weight management medications. The study in our laboratory shows anti-adipogenic potential of CQ solvent fractions and its implications on obesity. Analysis of cell viability, growth, proliferation and metabolic activity assays on 3T3-L1 cell line revealed the non-cytotoxic, metabolic and proliferative concentrations of Ethyl Acetate (CQ-EA) and Butanol (CQ-B) (0.0001µg/ml, 001µg/ml, 1 µg/ml and 10 µg/ml). 3T3-L1 cells were treated with these non-detrimental doses of selected solvent fractions during cell differentiation into adipocytes (adipogenesis). In correlation with its results, expression profile analysis was also performed which revealed that CQ fractions downregulated expression of adipogenic and lipogenic marker genes including PPAR-γ, aP2, LPN and ADIN. The expression profiles of adipogenic marker and lipogenic marker genes indicate that these CQ solvent fractions (CQ-EA and CQ-B) have anti-adipogenic potential. This conclusion correlates with the results of triglycerides estimation and semi-quantification of neutral lipids by ORO staining. The threshold for a good anti-obesity drug was set at reduction of 40 – 50% triglycerides and neutral lipid content of the cells. Cissus quadrangularis proves to be responsible for inhibition or delay in the cell differentiation process (3T3-L1) and lipid synthesis, acting as a good anti-obesity drug which should also enhance synthesis of cytokines (adiponectin and leptin).

#### 3. HUMAN AND ANIMAL DISEASES

# DEVELOPING INNOVATIVE AND ALTERNATIVE REGIME FOR CONTROLLING BOVINE TUBERCULOSIS IN ANIMAL-HUMAN INTERFACE

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Bovine tuberculosis (bTB) is a neglected endemic zoonosis, causing lot of mortalities every year. Previous methods of controlling bTB, like vaccination, antibiotics, have raised the concern of animal product consumers regarding microbial resistance. These measures have not been proved effective in developing countries, so disease prevalence in these areas is increasing continuously. In a report from WHO, Pakistan have been declared Eight out of ten countries with the highest incidence of bTB. So, there is dire need to find new therapeutic and preventive measures against Mycobacterium infection. Identification of selection signatures for genetic resistance is promising new alternate to combat bTB. Many of countries have opted this new approach and have reported useful data in cattle but very limited efforts have been put into the river buffaloes. River buffaloes of Indo-Pak region are world famous for their superior genetic potentials and inter-breed variations that provides substantial basis for identification of significant selection signatures. Present research was planned to explore IFNg and Toll-Like receptor genes in river buffalo for its association with bTB. Interferon gamma (IFNg) and Toll-Like receptor genes are key responder cytokines in Mycobacterium infection. For their genetic characterization, blood was collected from tuberculin negative (n=267) and tuberculin positive (n=194) animals. DNA was extracted and Sanger's method of DNA sequencing was used. Significance of each variation was tested by Hardy Weinberg equilibrium (P<0.05). Association was performed by one way ANOVA. Sequence comparison of two groups provided a total of five variations. Results illustrated only one variation found significantly associated with better immunity against bTB. For purpose of some additional and supportive information phylogenetic analysis was also being performed by neighbor joining method with bootstrap value-1,000. Tree indicated that river buffaloes are in closest proximity to Bos taurus and its genetic distance from other species may also be seen in Figures provided.

# SEROPREVALENCE OF BORRELIA BURGDORFERI SENSU LATO IN CAMEL (CAMELUS DROMEDARIUS) IN PUNJAB, PAKISTAN

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A seroprevalence study conducted on the presence of *Borrelia burgdorferi* sensu lato antibodies in camel of two district of Punjab, Pakistan from May 2019 to January 2021. The aim of present study was to detect the antibodies against *Borrelia burgdorferi* sensu lato in camels. A total of 405 serum samples were collected from two districts of Punjab i.e., Bhakkar and Bahawalpur. A questionnaire was used to collect data regarding potential risk factors like gender, age and tick infestation. Serological examination revealed the positive percentage of *Borrelia burgdorferi* sensu lato in camels was 2.47% (10/405). Risk factor analysis showed that gender, age and tick infestation are significantly (p < 0.05) associated with occurrence of borreliosis in camels. This study reports, for the first-time, presence of antibodies against *Borrelia burgdorferi* sensu lato in camels in Pakistan. Camels may play an important role in the transmission of borreliosis in other animal species as well as humans in Pakistan.

# ASSESSMENT OF RESPIRATORY PROBLEMS IN WORKERS ASSOCIATED WITH INTENSIVE POULTRY FACILITIES IN PAKISTAN

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The poultry industry in Pakistan has flourished since the 1960s; however, there are scarce data regarding the impact of occupational exposure on the pulmonary health of farm workers in terms of years working in the industry. The objective of the present study was to assess the effect of poultry environment on the health of occupationally exposed poultry farmers in countries of warm climatic regions, such as Pakistan. This study will also show the effect of exposure to poultry facilities on the health of poultry farmers in the context of low-income countries with a relatively inadequate occupational exposure risk management. The lung function capacity of 79 poultry workers was measured using a spirometer. Along with spirometry, a structured questionnaire was also administrated to obtain information about age, height, weight, smokers/nonsmokers, years of working experience, and pulmonary health of farm workers. The workers who were directly involved in the care and handling of birds in these intensive facilities were considered and divided into four groups based on their years of working experience: Group I (3-10 months), Group II (1-5 years), Group III (6-10 years), and Group IV (more than 11 years). The forced vital capacity (FVC), forced expiratory volume in one second (FEV1) and the FEV1/FVC ratio were considered to identify lung function abnormalities. Statistical analysis was carried out using independent sample t test, Chi-square test, Pearson's correlation, and linear regression. Based on the performed spirometry, 68 (86 %) of workers were found normal and healthy, whereas 11 (14 %) had a mild obstruction. Of the 11 workers with mild obstruction, the highest number with respect to the total was in Group IV (more than 11 years of working experience) followed by Group III and Group II. Most of the workers were found healthy, which seems to be because of the healthy survivor effect. For the independent sample t test, a significant difference was noticed between healthy and nonhealthy farmers, whereas Chisquare test showed a significant association with height, drugs, and working experience. Linear regression that was stratified by respiratory symptoms showed for workers with symptoms, regression models for all spirometric parameters (FVC, FEV1, and FEV1/FVC) have better predictive power or R square value than those of workers without symptoms. These findings suggest that lung function capacity was directly related to years of working experience. With increasing number of working years, symptoms of various respiratory problems enhanced in the poultry workers. It should be noted that most of the poultry workers were healthy and young, the rationale being that there is a high turnover rate in this profession. The mobility in this job and our finding of 86% of the healthy workers in the present study also proposed healthy worker survivor effect.

#### ASSESSMENT OF TUBERCULOSIS IN PATIENTS VISITING DISTRICT HOSPITAL CHARSADDA

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Tuberculosis is an infectious disease caused by rod shape bacteria called mycobacterium. Tuberculosis generally affects lungs, but can also affect other parts of the body such as bones, lymph organs and intestine. Tuberculosis is more common in people with immune system problem, the classic symptoms of active TB are a chronic cough, fever, night sweats and weight loss. The study aimed to found out the prevalence tuberculosis in District Charsadda, Khyber Pakhtunkhwa. The study was conducted at DHQ hospital Charsadda. The Secondary data was used from hospital record. A total of (921=100%) patients were screened to investigate the presence of TB bacteria during three month (). The Majority of people of age 25-45 were suffering from tuberculosis. The prevalence was higher in males than female. According to the present study the ratio of pulmonary tuberculosis is higher than extra-pulmonary tuberculosis in district Charsadda. The patients of pulmonary tuberculosis were (500=54%) and extra-pulmonary was (421=45%).

# RISK FACTORS ASSOCIATED WITH AN OUTBREAK OF DENGUE FEVER IN DISTRICT KASUR, PUNJAB, PAKISTAN

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Dengue fever (DF) appeared to be emerging in 112 union councils of District Kasur Punjab, Pakistan in January to November in 2021. A controlled case study was performed to investigate risk factors for the surveillance of DF in Tehsil Kasur (43 union councils), Pattoki (27 union councils), Kot Radha Kishan (11 union councils) and Chunian (31 union councils) of district Kasur. A total of 132 patients with DF were studied in all the union councils of all four tehsils of the district Kasur. Out of 132 patients 96 were male and 36 were female. We further divided under study patients in three categories such as below 25 years (38 patients), 25 to 40 years (63 patients) and above 40 years (31 patients). The risk factors for these patients were factories, water ponds, godowns, swimming pools, nurseries, service stations, abundant buildings, marriage halls, hotels, schools, grave yards, mosques, religious places, parks, tyre shops, junk yards, workshops, railway stations, colleges, bus terminals, tube wells, grid stations, high raised buildings, parking stands, horticulture places, filtration plants, dairy plants, open sewerage system, supply of water through water tanks, its improper storages at homes, unhygienic conditions found during indoor survey especially in rented areas, under construction buildings and living in a house discharging sewage directly into to ponds were all significantly associated with DF. These all risk factors provided best habitat for the growth of dengue larva at these areas. These results contributed to the understanding of the dynamics of dengue transmission in 112 union councils from which 48 were red (with dengue patients and dengue larva), 56 yellow (with dengue larva) and 8 were green (neither dengue patients and dengue larva) and its vicinity, which was needed to implement dengue prevention and control programmes effectively and efficiently.

# IDENTIFICATION AND CHARACTERIZATION OF SESQUITERPENE LACTONES AS POTENTIAL FALCIPAIN-2 INHIBITORS

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Malaria is a disease transmitted through the bite of a female Anopheles species of mosquito infected with one of the four species of plasmodium: Plasmodium falciparum, Plasmodium vivax, Plasmodium ovale and Plasmodium malariae in humans. Plasmodium falciparum is a protozoan parasite that causes the most virulent form of human malaria and kills at least one million children annually. According to World Health Organization (WHO) reports 1999–2004, over two billion people, representing more than 40% of the world's population, are at risk of contracting malaria, and the number of malaria deaths worldwide has been estimated to be 1.1-1.3 million per annum. In Pakistan, 500,000 malaria infections and 50,000 malaria-attributable deaths occur each year. At present, different anti-malarial drugs are available in the market for example, Chlorquine and other similar Quinolones (e.g. Hydroxychloroquine), Quinine, Primaquine, Mefloquine, Sulfonamides and Artesunate & Artemether (Artemisinin analogs). However, Plasmodium falciparum has developed resistance to all these drugs. Falcipain-2 (FP-2) of Plasmodium falciparum is a papain-family (C1A) cysteine protease that plays an important role in the parasite life cycle by degrading erythrocyte proteins, most notably hemoglobin. Inhibition of FP-2 and its paralogues prevents parasite maturation, suggesting these proteins may be valuable targets for the design of novel anti-malarial drugs. Since prevailing anti-malarial drugs are suffering from drug resistance, therefore identification of new novel bioactive molecules with potent anti-malarial activity are highly desirable. Here in this study, we found two sesquiterpene lactones; Alantolactone and Brevilin A as falcipain-2 inhibitors. Alantolactone has been found to show hydrophobic interactions with TRP206, CYS42 and HIS174 residues of 2GHU at a bond distance 3.69181Å, 4.70387Å and 5.28753Å, respectively. The binding energy value obtained from the interaction of Alantolactone with 2GHU was -7.2kcal/mol. The inhibition constant value acquired from binding energy value was 5.205μM. While our second docked compound Brevilin A form hydrogen bonding with ASN173 and TRP206 residues of 2GHU at a bond distance 2.40334Å and 2.50892Å, respectively. The binding energy value obtained from the interaction of Brevilin A with 2GHU was -8.1kcal/mol. The inhibition constant value acquired from binding energy value was 1.136μM. Our Molecular Docking study results have suggested that both studied sesquiterpene lactones are potential inhibitors of falcipain-2 but Brevilin A is predicted to be the best inhibitor of falcipain-2 as it forms strong bonding (hydrogen bonding) with amino acid residues of falcipain-2 and has lower value of binding energy and inhibition constant. Further *in vivo* and *in vitro* studies should be done to validate these findings and develop these two bioactive sesquiterpene lactone compounds into novel anti-malarial drugs.

# ASSOCIATION OF EPIDEMIOLOGICAL AND HEMATOLOGICAL PARAMETERS WITH REPEATED SPONTANEOUS MISCARRIAGES DURING FIRST-TRIMESTER OF PREGNANCY: A CASE CONTROL STUDY

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We aimed to investigate the association of first trimester repeated spontaneous miscarriages with hematological profile, blood group and age. This case-control study was conducted at the laboratories of Zoology department The Women University Multan and the Department of Hematology, Nishter Hospital Multan, from June 2016 to Dec 2019. A total of 500 participants, 300 with spontaneous abortion and 200 healthy pregnant controls in first trimester of pregnancy were included in this study. The data was collected through questionnaire at the time of sampling after obtaining informed consent. ABO and Rh blood group was identified by using Antisera (including Anti-A, Anti-B and Anti-D) The analysis of haematological indices was done by using automated haematological analyzer (Sysmex Co. Japan). The mean maternal age in this study was 30.35±7.94 years for cases and 28.53±6.59 years for controls. Highest frequency of miscarriages 90 (30%) was observed in cases of age group 35-39 while lowest frequency of miscarriages 30 (10%) was observed in age group 20-24. Whereas, highest frequency 94 (47%) and lowest 2(1%) of controls were observed in age group 25-29 and 15-19 respectively. Out of 500 women 438 (87.6%) were Rh positive (including 256 cases and 182 control) and 62 (12.4%) were Rh-negative (including 44 cases and 18 controls). The observed ABO distribution pattern was B>A>O>AB. Blood group B was most frequent (34%) including 18% cases and 16% controls. Percentage of Rh-negative was higher in cases than controls 14.6% vs 9% but ABO and Rh blood groups showed no association with RSM in this population p=0.372; p=0.08 respectively. Haematological evaluation revealed significant decrease in Hb (p<0.001), MCV (p<0.001) and MCHC (p=0.006) values in cases as compared to controls whereas no significant differences found between the case and control groups in terms of RBC, WBCs, HCT, MCH, neutrophils, lymphocytes, and PLT (p>0.05). It is concluded that the age groups 35-39 years and significant changes in the values of Hb, MCV and MCHC may be associated with repeated spontaneous miscarriage in this population.

# A DESCRIPTIVE EPIDEMIOLOGICAL STUDY OF HEAD AND NECK CANCER IN NISHTAR HOSPITAL MULTAN

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Number of head and neck cancer patients is increasing every day worldwide. Much research has been done all over the world regarding different features of HNC but in Southern Punjab little or no work has been done. The purpose of this study was to find out the prevalence and clinicopathological features of Head and Neck Cancer

(HNC) patients visiting Nishtar Hospital Multan. Data was collected from biopsy proven Head and Neck Cancer patients during April 2017 to July 2019 and analyzed for clinico-pathological features. The ratio of HNC was more in male patients as compared to female patients Mean age of patients was 54.77±15.96 and 44.31±14.79 for male and female respectively. Majority of the patients belonged to Dera Ghazi Khan Division. The habit of smoking was identified in 51% patients and 49% patients were nonsmokers. Majority patients belonged to poor family background and vegetarians were more frequent. Only 47.1% patients did care of oral cleanliness. More than half of the patients were of stage III. Moderately differentiated tumour (Grade 2) was the most frequent grade. Regarding tumour size and nodal status, T3-4 and N0 were more frequent with frequency of 71.0% and 50.4% respectively. The Larynx was found to be the most common affected site of HNC patients with significant subsites of Glottic area. Squamous cell carcinoma was the commonest type as compared to Basal Cell Carcinoma, Adenocarcinoma and Lymphoma.

#### SEROPREVALENE OF TOXOPLASMOSIS IN RESIDENCE OF KAROR PAKKA DISTRICT LODHRAN

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This study was conducted to evaluate the prevalence of toxoplasmosis in humans of Kehror Pakka, district Lodhran. The blood sample (n=92) were collected in EDTA vacutainers, and then prevalence of toxoplasmosis was analyzed through commercial Enzyme Linked Immunosorbent Assay (ELISA) kit and also studied the effect of Toxoplasma gondii on haematology of hosts. The overall prevalence of toxoplasmosis was 45.16%, the relationship between gender of peoples revealed that prevalence was high in females (45.16%) than males (45%). Similar relationship between the age groups 12-23, 24-35, 36-47 and >48 years showed prevalence 48.14, 39.53, 46.66 and 62.5% respectively, which showed higher prevalence in age year >48 and lower in age year 24-35. The relationship between literacy rate of peoples indicated that prevalence was higher in non-educated males (45.5%) than educated males (44.4%). Similarly, prevalence was high in educated females (57.14%) than non-educated females (37.5%). The relationship between socioeconomic status and prevalence of toxoplasmosis showed 48.57, 40 and 62.5% in poor, middle and rich peoples respectively. For analysis of hematological parameters blood samples were collected. The Mean  $\pm$  SEM values of WBC, LYM, MID, GRA, HGB, MCHC, MCH, and MCV were  $8.731 \pm 0.609$ ,  $2.914 \pm$  $0.246, 0.7078 \pm 0.0493, 7.03 \pm 2.03, 9.748 \pm 0.449, 28.868 \pm 0.431, 24.051 \pm 0.790,$  and  $82.86 \pm 1.85$  respectively high in non-infected peoples. Whereas RBC, RDW-SD, RDW-CV, HCT, PLT, MPV, PDW, PCT and PLCR were  $4.2910 \pm 0.0921$ ,  $46.751 \pm 0.611$ ,  $17.227 \pm 0.570$ ,  $34.45 \pm 1.02$ ,  $271.9 \pm 35.1$ ,  $8.437 \pm 0.164$ ,  $12.842 \pm 0.406$ , 0.2235 $\pm$  0.00258, and 27.62  $\pm$  1.53 respectively high in infected peoples.

#### PREVALENCE OF GIARDIA LAMBLIA IN CHILDREN OF PESHAWAR

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Giardia lamblia is a water-born protozoan that infects intestinal tract causing infectious gastroenteritis. In developed countries, Giardiasis account for 2-7% of all diarrheal illness. Clinical manifestation of Giardiasis can be variable ranging from asymptomatic to acute or chronic diarrheal diseases. The present study was conducted to determine the age, sex and month-wise prevalence of Giardia lamblia in children aged 1-12 years frim April to September 2018. A total of 150 children were enrolled in the study. Stool samples were collected and analyzed for parasite presence using microscopy. Out of 150 samples Giardia lamblia was detected in 6 (4%) cases. Prevalence was higher in children with aged 5-8 years (6.1%) compared to other groups. Male children were more infected

(4.2%) as compared to female children with a Prevalence rate of (3.2%) respectively, but the difference was not significant. Different seasons also affected the Prevalence rate of Giardia lamblia being more prevalent in summer. The higher Prevalence rate of 11% was observed in the month of August followed by 8.3% and 4.3% in the months of July and September respectively. The lowest rate was recorded in month of April 2018 and September 2018, 150children were randomly selected. A stool sample with socio-demographic data was collected from each child. Climate change and population growth are also predicted to increase both malnutrition and the prevalence of these parasites in children.

#### 4. MICROBIOLOGY

# ANTIMICROBIAL EFFECT ON COMFORT PROPERTIES (ABSORBENCY, AIR PERMEABILITY) OF COTTON FABRIC

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Cellulosic fabrics have great surface area and ability to provide good atmosphere for the progression of microbes thus they produce bad smell and irritation problems beside with textile weakening. This study was aimed to applying eco-friendly antimicrobial finish on textiles. The antimicrobial finish was extracted from leaves of *Azadirachata indica, Butea monosperma* and *Litchi chinensis* plants and applied on 100% cotton. Before and after applying, presence of microorganisms, FTIR, SEM, fabric properties and sustainability to washes were checked. The antimicrobial finish was applied by pad dry cure method and finish was fixed by using of poly urethane binder. The presence of microorganisms was checked by ASTEM E2149 shake flask method. The results were analyzed through MANOVA and ANOVA. The fabric properties were checked by using AATCC and ISO standard test methods. The eco-friendly antimicrobial finish made 89% reduction in microbial growth. In case of comfort related properties (Absorbency, Air Permeability) antimicrobial finish had positive effect on cotton fabric. The antimicrobial finish lasted up to 25 washes. The study suggested that antimicrobial fabric is suitable to provide protection cover for medical industry, paramedical staff, sports wears, home furnishing as well as common people.

# EVALUATION OF METHICILLIN RESISTANCE IN FIELD ISOLATES OF *STAPHYLOCOCCUS*AUREUS: AN EMERGING ISSUE OF INDIGENOUS BOVINE BREEDS

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The emergence of resistant strains of *Staphylococcus aureus* (*S. aureus*) particularly methicillin-resistant *S. aureus* (MRSA) confers overwhelming economic losses to the global dairy industry. The current study was planned to investigate the *S. aureus* and MRSA associated subclinical mastitis in 345 milk samples (Cattle n=173, Buffalo n=172) collected from indigenous bovines of district Rawalpindi. The milk samples were screened for *S. aureus* and the confirmed isolates were subjected to disc diffusion test, PCR and SDS-PAGE analysis for the confirmation of methicillin resistance. The results revealed an overall molecular prevalence of 28.69% for *S. aureus among which* MRSA-associated mastitis was found 47.61% prevalent. The SDS PAGE analysis depicted the presence of a 78KDa protein band specific for *PBP2a* protein in MRSA. The comparative risk factor analysis showed significant variation among risk factors associated with *S. aureus* and MRSA-induced mastitis. The phylogenetic analysis of MRSA *mec*A gene showed a high resemblance of the study isolates with MRSA isolates of the USA, Turkey, India, Africa, and Brazil. This is the first study regarding the molecular characterization and phylogenetic analysis of MRSA isolates from study area.

### OPTIMIZING THE ANTIBACTERIAL EFFECT OF VINEGAR ON CHOPPED FRUITS AND VEGETABLES

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Pre- and post-harvest contaminants threaten the safety of fruits and vegetables. Sanitizing effect of vinegar on some fresh fruits and vegetables was assessed for consumer's satisfaction. Five samples of chopped fruits and vegetables i.e. cucumber, melon, carrot, banana and apple were purchased from locally super market in Shad Bagh Lahore, Pakistan. The samples were treated with 2.5%, 5%, 7.5% and 10% acetic acid for 0, 15, 30, 45 and 60 minutes. Samples treated with 7.5%and 10%, acetic acid showed bacterial inhibition in fruits and vegetable after 30 min, 45 min and 60 minutes, at 28 °C to 31 °C. This study shows that acetic acid can be used as an effective sanitizer for fruits and vegetables for the inhibition of microbial contaminants and their growth.

#### ASSESSMENT OF FRUIT BAT'S (PTEROPUS MEDIUS) ORAL MICROBIOTA THROUGH N.G.S

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A total of 5 specimens of *Pteropus medius* were captured from different roosting sites within the district Kasur, Punjab, Pakistan using mist nets. Sterile cotton swabs were used to collect saliva samples. DNA was extracted using QIAamp DNA Microbiome kit following the manufacturer's instructions. The next-generation sequencing of PCR products was done using overseas commercially available facility at macrogen Korea. The analysis result of bacterial species annotation is visually shown in KRONA plot. The abundance of identified bacterial phylum was Proteobacteria 81% > Firmicutes and Actinobacteria were 16%. Bacterial classes include Bacteroidia, Saccharimonadia, Negativicutes, Actinobacteria, Bacilli and Gammaproteobacteria were identified. The order of abundance was Gammaproteobacteria 81% > Bacilli 15% > Actinobacteria 3%. Similarly, Saccharimonadales, Xanthomonadales, Betaproteobacteriales, Pseudomonadales, Selenomonadales, Pasteurellales, Corynebacteriales, Bacillales, Lactobacillales were different bacteria order identified in saliva of fruit bat. The relative abundance of identified families was Enterobacteriaceae 79% > Streptococcaceae 8% > Corynebacteriaceae 3% > Pasteurellaceae 1%. Bacteria genera including Actinobacillus, Pseudomonas, Veillonella, Weissella, Pantoea, Staphylococcus, Streptococcus and Escherichia were identified. The percentage of Escherichia was 76%> Streptococcus 8%> Staphylococcus 5% > Pantoea 2% > and Weissella were 1%. The relative abundance of Escherichia Shigella was 76% followed by Staphylococcus aureus which is 3%. In our findings it can be concluded that fruit bat (Pteropus *medius*) can be the reservoir of pathogenic bacteria and can affect directly humans and livestock.

## BACTERIAL LACCASES MEDIATED DECOLOURIZATION AND DEGRADATION OF DIANIX YELLOW BROWN DISPERSE

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Rapid industrialization poses a serious threat to both the environment and human health because of toxic effluents. The conventional physicochemical approaches are not efficient to eliminate all types of industrial effluents. The bioremediation of industrial waste can be made more efficient by using ligninolytic laccase enzymes. Bacterial laccases actually belongs to the multicopper oxidases family and is engaged in the crosslinking of monomers, which is important in the degradation of a variety of industrial pollutants. The present research was employed to assess the decolorization and biodegradation efficiency of Dianix yellow brown, a textile disperse dyes by using laccase

producing bacteria. Three locally isolated laccase producing bacterial strains namely GY3, AY4 and AY1 showed significant biodegradation activity against Dianix yellow brown. The decolorization of dianix yellow brown resulted change in color from yellow brown to light yellow color. Under optimum conditions biodegradation efficiency of dianix yellow brown by using laccase producing bacterial strains was 72% with GY3, 89% with AY1 and 91% with AY4 after 48h. UV Visible spectroscopy and Fourier-transform infrared spectroscopy (FTIR) confirmed the biodegradation of dianix yellow brown.

## PREVALENCE OF MULTIPLE-METAL-RESISTANT BACTERIA IN AIR COLUMN AT DIFFERENT LOCALITIES OF LAHORE, PAKISTAN

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The study has been carried out to check out for the prevalence of various metal resistant bacteria in air column at Lahore, Punjab, Pakistan. Six different metals (Cu, Cr, Ni, Pb, Zn and Fe) have been used during this study, as the number of bacteria tolerant to each specific metal is checked at four different concentration levels (250ppm, 500ppm, 750 ppm and 1000 ppm). Load of aforementioned heavy metal resistant bacteria was observed from ten separate localities of Lahore district (Thokar Bypass, Thokar Niaz Baig, Doctor Hospital Under pass, Campus Under pass, Muslim Town Under pass, Dharampura Underpass, Mughalpura Under pass, Pakistan mint, Daroghawala Chowk and Iqbal colony). Bacterial colonies of Lead, Iron, Copper and Nickel resistant bacteria were clearly observed while Chromium and Zinc don't have any bacterial growth. Thus, representing the much higher proportion of air pollution in Lahore district, caused by Pb, Fe, Cu and Ni whereas comparatively much lesser air pollution prevailing due to Cr and Zn. Three localities namely Daroghawala Chowk, Iqbal colony and Muslim Town Under pass are under the worst scenario in case of air pollution caused by these observed heavy metals because of industrial waste (Daroghawala Chowk and Iqbal colony) and waste from heavy traffic (Muslim Town Under pass).

# EFFICACY OF HONEY AS ANTIBIOFILM, ANTIQUORUM SENSING AND DISPERSAL AGENT AGAINST MULTISPECIES BACTERIAL BIOFILM

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Bacterial biofilms are a major worldwide healthcare problem (urinary tract infections) and are associated with decreasing quality of life and significant patient morbidity. This study is first to test Pakistani honey bess, Apis dorsata and A. cerana honey samples as anti biofilm, anti quorum sensing (QS) and biofilm dispersal agents honey against multispecies biofilm of bacteria (obtained from obese patients). Briefly, five previously identified isolates Pseudomonas aeruginosa, Escherichia coli, Staphylococcus aureus, Morganella morganii and Klebsiella pneumoniae (MT448672-MT448676) were selected. Antibiogram study of all five isolates was tested against three antibiotics viz., erythromycin (20 µg/mL), lincomycin (100 µg/mL) and rifampicin (100 µg/mL). In order to form multispecies biofilm, identified bacteria were grown in batch culture by mixing equal volumes (OD<sub>590nm</sub>= 0.1) of 2, 3 and 5 bacterial isolates. In total 11 groups (g1-g11) were made. Crystal violet (CV) staining method was used to evaluate the antibiofilm potential and biofilm dispersal potential of both honey samples. QS inhibition in P. aeruginosa was measured following culture supernatant method. Antibiogram study showed significant (p < 0.05) resistance by P. aeruginosa against tested antibiotics. E. coli, M. morganii and K. pneumoniae were significantly susceptible to erythromycin and S. aureus to lincomycin. Minimum inhibitory concentrations (MIC) values of both honey samples showed 2 and 5% concentrations as having significant (p < 0.05) inhibition potential of multispecies biofilm by all test groups (g1-g11). Though A. dorsata honey significantly inhibited biofilm formation at 2 and 5% against all groups but 2% concentration was highly significant against g2-g4 groups. Regarding A. cerana honey, 2% concentration was significantly effective against g1, g4-g7 and g9-g11 groups. Both honey samples significantly inhibited QS at 2 and 5%. The 5% concentration of *A. dorsata* honey significantly dispersed biofilm by all groups compared to 2% which showed dispersal potential only by g2 and g3 groups. Collectively, honey samples showed significant antibiofilm, anti-QS and biofilm dispersal potentials thus can be considered as good alternative to antibiotics.

### ROLE OF DIRECT FED MICROBIALS (DFM) IN THE ENHANCEMENT OF NUTRITIONAL CAPABILITIES OF RUMINANTS- A CURRENT SCENARIO

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The gastrointestinal tractc (GI) is populated with as many as 2000 different types of bacteria. Some of these bacteria are beneficial, while others are pathogenic. The balance between these two types of bacteria helps the overall health of the animal. All bacteria require a source of nutrition for survival and beneficial bacteria and pathogenic bacteria compete for nutrients and attachment sites on the gastrointestinal mucosa. However some of the beneficial bacteria in the GI tract, such as bifidobacteria, use specific substrates for nourishment that pathogenic bacteria can not. Feeding a prebiotic that serve as a source of nutrition for the probiotic organism but not for the pathogenic bacteria gives a beneficial bacteria a competitive advantage over pathogenic bacteria. For instance, simultaneously feeding a probiotic bifidobacterium Animalis ACH7 with the prebiotic fructooligosaccharides (FOS) encourages the growth and survival of Bifidobacterium animalis ACH7. Feeding the prebiotic also results in increase persistence of probiotic in the GI tract. Probiotics are live microbial feed supplements which beneficially affect the host by improving its microbial intestinal balance. Correspondingly, in feed regulation probiotics are included in the group of feed additives stabilizing the microbial communities of the digestive tract in monogastric animal and ruminants. They are also known as digestive bio regulators or direct fed microbials. Well known examples are the use of bacteria mainly lactic acid bacteria (LAB) for production of silage, fermented cabbage and sour milk products such as yogurt, cottage cheese and kefir and the use of yeasts mainly Saccromyces cervicae for production of bread, beer and wine. As man and animals are born with a sterile digestive tract, but very soon after the birth a wide diversity of microorganisms begins to colonize the digestive tract. The digestive compartments which are the richest in microbes are the fore stomach (rumen) in polygastric animals while large intestine in the man and monogastric animal. An open and complex ecosystem is created which has an essential role for the host. On one hand the digestive microflora is involved in digestion, on the other hand it has a local impact on the immune system, thus offering the possibility to exert a positive and completely natural effect on health, well being and performance of animal through its autochotonous microflora. Supplementing different probiotics (fungi/yeast and bacteria) resulted in improved nutrient status and productivity of the ruminants under certain conditions. Lactic acid bacteria has been used for millennia in the production of fermented milk products and silage. Some form the main intestinal microflora and therefore an indispensable form of the resident microflora in man and animal. Lactic acid bacteria convert certain type of sugars by fermentation mainly into lactic acid. Important lactic acid bacteria in probiotics belong to the genera including Lactobacilli, Pediococci, Bifidobacteria and enterococci. Enterococcus faecieum previously known as Streptococcus faecium is the most important species used in the animal nutrition. More investigations are needed with different diets to confirm these effects and improve the knowledge on the mode of action of BL as additive for ruminants.

#### PREVALENCE OF SUPERFICIAL HUMAN PATHOGENIC FUNGI IN, GILGIT

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Fungi are eukaryotic unicellular such as yeasts to multicellular organisms such as puffballs with cell wall composed of chitin. They are fundamental for life on earth in their roles as symbionts, e.g. in the form of mycorrhizae, insect symbionts, and lichens. The total number of eukaryotic species on Earth has recently been estimated at 8.7 million with fungi making up approximately 7% (611,000 species) of this number. The fungal lineage is one of the three large eukaryotic lineages that dominate terrestrial ecosystems. They share a common ancestor with animals in the eukaryotic super group Opisthokonta. Fungi are of an ancient lineage and have a fossil record that extends back to the Devonian and Pre-Cambrian era. The earliest written record of fungi are not of the fungi themselves, but of their depredations. To the physician and poet Nicander fungi were 'the evil ferment of the earth; poisonous kinds originating from the breath of vipers. Mycology is that branch of biology where we studying about fungi. Fungi are multicellular eukaryotic fungi ranging from unicellular to multicellular fungi like yeasts. They are fundamental for life on earth in their roles as symbionts, e.g. in the form of mycorrhizae, insect symbionts, and lichens. Likewise, fungi have its own significance regarding its beneficial to harmful effect ever on plants, animals or humans. Human pathogenic fungi is more prevalent in our environment due its consequences on human's hiddnley that's why called as "Hidden killers". Medical Mycology, a study of fungal epidemiology, ecology, pathogenesis, diagnosis, prevention and treatment in Human beings, is a newly recognized discipline of biomedical sciences, advancing rapidly. Fungal infections today are among the most difficult diseases to manage in humans. These diseases kill more than 1.5 million and affect over a billion people. Fungal diseases are a worldwide problem ranging from superficial infections easy to cure to more invasive life-threatening infections that are much harder to diagnose and treat. Recent estimates suggest that invasive fungal infections cause at least as many deaths as malaria and tuberculosis. The burden of fungal disease continues to increase as the number of people with weakened immune system increase. They attack people with serious illness, and frequently jeopardize the success of the newest medical advances in cancer care, solid organ and hematopoietic stem cell transplantation, neonatal medicine, autoimmune disease therapies, trauma and intensive care, and sophisticated surgery. Gilgit city is the main city of GB, which is the main attraction area to people for its beauty and resource purposes like education sectors, job opportunities and health facilities. People use together for different purposes related to job, study or other work. This city is mainly surrounded by other towns like danyore, Shanglidaar and oshikhnadass. There are 4 main hospitals found in Gilgit are; DHQ hospital, City hospital, GMC hospital and CMH hospital Gilgit. Along with these hospitals one newly build hospital consist of 30 beds known as Shanglidaar hospital located in between Muhammad Abad danyore and oshikhnadass. According to my area of research 3 hospitals were chosen to collect human fungal skin samples are; DHQ hospital, City hospital and Shanglidaar hospital. Total of 30 patients were examined collectively from these hospitals, 14 fungal species (Cryptococcus Neoformans, Actinomyces Israelii, Ajellomyces, Candida albicans, Cladosporium, Coccidioides ssp, Cryptococcus Neoformans, Emmonsia ssp, Geotrichum Candidum, Lacazia Loboi, Microsporum audouinii, Microsporum canis, Malassezia Fur Fur and Trichophyton Rubrum). The identified ssp belonging to 8 families and 10 genera the highest number of ssp is from Tinea which is four in number of genus Trichophyton. Identification was done in KIU biology Lab.

#### LOW COST PRODUCTION OF BACTERIAL EXTRACELLULAR POLYSACCHARIDES

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Various industries manufacture polymers for well beings of human from centuries. But the industrial polymers have less uses in numerous fields for human as compared to the biopolymers. The exopolysaccharides or extracellular polysaccharides (EPS) are the biopolymers produced from cells. Both microbial and plants cells produce the extracellular polymers. But the plants biopolymers took long time for their generation and have less carbon chain

length, molecular weight as well as less branching as compared to the microbial polymers. The high degree of branching and high molecular weights of EPS has many uses in the various fields of life, especially in pharmaceutical industries. This study mainly emphasized on high molecular weight exopolysaccharide production through microbes using low-cost substrates. For this purpose, pure strain of halophilic bacterium *Bacillus licheniformis* JF38 was grown on sucrose containing medium as well as on the low-cost substrates like, potato peel, grape fruit peel and sugar cane bagasse. The product formed was tested by specific rotation, paper chromatography, TLC and FTIR. This study concluded that the one of these low-cost substrates i.e potato peel was equally effective for production of levan exopolysaccharide as that of expensive substrate like sucrose.

#### 5. PHYSIOLOGY

#### TRACE ELEMENT COMPOSITION IN WHOLE BLOOD OF PREGNANT WOMEN

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Elements like calcium (Ca), magnesium (Mg), iron (Fe), zinc (Zn), copper (Cu), cobalt (Co), nickel (Ni), chromium (Cr), manganese (Mn) and cadmium (Cd) are extremely important in maintaining the metabolism in human body. During pregnancy the metabolic demand of mother increases to support the metabolism of developing fetus and as a result the micronutrient demand of a mother's body increases. Micronutrient deficiencies may cause significant risks during pregnancy like pregnancy induced hypertension, gestational diabetes mellitus, anemia, preeclampsia, miscarriage or labor complications. Although trace metal concentration was detected in general but the relationship of trace elements to the etiology of pregnancy is yet to be identified. . This study focused to estimate the levels of micro and macronutrients in whole blood of Pakistani pregnant women to investigate the changes in the concentration of these elements with the progression of gestation. Approval of study was obtained from "Bioethics Committee for research on Human Subjects", Faculty of Biological Sciences, Quaid-i-Azam University, Islamabad, Pakistan. A study group of 40 healthy non diabetic, non-hypertensive females was recruited and further divided into two age groups (Group I:20-30 years and Group II:30-40 years). Another group of 20 healthy non-pregnant females was formed and further sub divided into two according to age. Their sociodemographic, anthropomorphic and diet record was maintained and blood was collected by puncturing antecubital vein. Samples from the study group were collected three times (1st, 2nd and 3rd trimester). Samples were predigested by adding conc. nitric acid (65% extra pure) and then manually digested on a hotplate at 400 °C inside a fume hood for the estimation of concentration of trace metals through flame atomic absorption spectrophotometry (FAAS). One way ANOVA was applied on data to compare the trace element concentration between study group and control group and between 1st, 2nd and 3rd trimester. Correlation was determined by Pearson's correlation and p<0.05 was considered as a statistically significant difference. The concentration of Fe in both groups was observed to decrease significantly in 2<sup>nd</sup> and 3<sup>rd</sup> trimester as compared to 1st and control group. Zn concentration was significantly lowered in 2nd and 3rd trimester as compared to non-pregnant control and 1st trimester in both groups. Ca level was significantly lowered as compared to 1st and control in group I, while in group II, the significant decrease in 2nd and 3rd trimester from 1st was observed. Mg in group I was significantly decreased in 1st, 2nd and 3rd trimesters as compared to control and also significantly lowered in 3<sup>rd</sup> from 1<sup>st</sup> in both groups. In both the groups, Mn levels were remained non-significantly different in 2<sup>nd</sup> and 3rd trimesters as compared to control and 1st trimester. Cu level was observed to be significantly elevated in 3rd trimester as compared to control and 1st in group I, while in group II, Cu was significantly increased in 3rd as compared to 1st and 2nd trimester. In group I, Co was also found to significantly increase in 2nd as compared to 1st and also in 3<sup>rd</sup> in comparison to control, 1<sup>st</sup> and 2<sup>nd</sup>. In group II, Co was also increased significantly in 3<sup>rd</sup> trimester as compared to 1st. Cr level was significantly lowered in 2nd and 3rd trimester as compared to 1st and control in group I. In group II, the level of Cr was also significantly lowered in 2<sup>nd</sup> and 3<sup>rd</sup> trimester as compared to 1<sup>st</sup>. In both groups, Ni was significantly lowered in 2<sup>nd</sup> trimester as compared to control and 1<sup>st</sup> while the level of Ni was significantly reduced in 3<sup>rd</sup> as compared to control. It has been observed that during the course of gestation Zn has a positive correlation with Ca and Mg. The correlation between Fe and Mg becomes strong in 2<sup>nd</sup> trimester and very strong in 3<sup>rd</sup> trimester. Ca and Mg positive correlation is the strongest of all during gestation becoming stronger by each passing trimester. The current study was focused on changes in the levels of trace elements in pregnant women during the course of gestation. The levels of trace elements show alteration in each trimester showing a gradual increase in Cu and Co levels but decrease in Fe, Ca, Mg, Cr, Ni and Zn levels. Concentration of Mn remained nonsignificantly different. Further studies are required to justify the levels of ultra-trace elements in pregnancy. Our findings will provide a great help in monitoring the maternal health status and fetal growth. However, further detailed and comprehensive studies are still needed in this regard. Although the study has provided a data on whole blood levels of trace elements concentrations of Pakistani women. There are, however, some limitations that must be noted. Firstly, the concentrations of some ultra-trace elements such as Co and Cr are at very low levels, thus the risk of contamination during blood collection, sample preparation and determination has to be seriously considered. Secondly, we have recruited only healthy females having no major disease, hypertension or diabetes mellitus but in future more studies can be performed recruiting pregnant females and comparing the levels of trace elements in diseased and healthy.

### DISORDERS ASSOCIATED WITH FEMALE INFERTILITY IN DISTRICT BAGH AND PONCH AZAD JAMMU AND KASHMIR

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The purpose of this study was to identify various disorders associated with female infertility and to calculate the frequency of these disorders. Data was collected by directly approaching known patients and by visiting different hospitals of Bagh and Rawlakot. A well-designed questionnaire was completed by patients. Out of 106 participants, 58 (54.7%) were found to have ovulatory dysfunction; 37 patients (34.9%) were found to have ovarian functional problems; 9 (8.5%) were found to have unexplained infertility; 8 patients (7.5%) were found to have tubal factors; 6 patients (5.7%) were found to have leiomyomas or fibroids; 4 (12.5%) secondary infertile women had preeclampsia; 2 (2.7%) primary infertile patients had PID and 4 patients (5.4%) with primary infertility had endometriosis. Various risk factors were found to be associated with female infertility including stress (67%), allergy (20.7%), liver and stomach problems (6.6%), tuberculosis (4.7%), thyroid problems (4.7%), hypertension (20.7%) and diabetes (5.6%). Ovulatory dysfunction was found to be the most prevalent disorder following ovarian functional problems which are linked with ovulatory dysfunction. The main causes of ovulatory dysfunction might include PCOS, sedentary lifestyle, obesity, and unhealthy eating habits. Lifestyle modifications can help to overcome these disorders. Moreover, there is a lack of awareness among females regarding health and fitness, steps must be taken to spread awareness and to educate females in this regard.

#### EFFECT OF GRADED DOSE OF VITAMIN E ON BLOOD AND SERUM BIOCHEMISTRY OF SHEEP

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Vitamin E is part of the non-enzymatic body's antioxidant system which fights with oxidative stress. Vitamin E defends cell membranes from oxidation through responding with lipid radicals which is formed in lipid peroxidation responses. The present study was conducted at livestock experimental Farm, department of Veterinary and medicine, University of Poonch Rawalakot of AJ&K to investigate the effects of vitamin E on the blood and serum biochemistry of sheep. For this study, all the sheep were weighed and divided randomly into three groups; Control, T1 and T2. T1 and T2 group were treated orally with two doses of vitamin E (150mg/kg body weight and 200mg/kg body weight respectively). At the end of the experiment, the results showed significant increase (P<0.05) in blood parameters in the vitamin E supplemented groups including RBC, RDW%, WBC, LYM concentration and LYM% while HCT%, HGB concentration, MCV, MPV, MCH, MCHC, MID concentration, MID%, GRAN concentration and percentage of GRA showed insignificant effect in treated groups. In serum biochemical parameters including albumin and AST concentration increased (P<0.05) in treated group while total protein and globulin concentration insignificantly effected by vitamin E in treated groups. So it is concluded that vitamin E had positive effect on the life of sheep.

### MENSTRUAL DISORDERS AND COMPLICATIONS OF DIFFERENT AGE GROUPS FROM THE MAIN HOSPITALS OF DISTRICT HYDERABAD AND JAMSHORO

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Menstrual disorder has been increasing over the entire world in both married and unmarried females. Menstrual disorder is physical and emotional problem that interfere with normal menstrual cycle causing pain and unusually heavy or light bleeding delayed menarche and missed period. Menstrual disorder is disruptive state in which the woman menstrual cycle becomes irregular. In the research project menstrual disorder was done by study of 20 patients from Liaquat University of Medical and Health Sciences, Jamshoro and civil hospital of Hyderabad during July to November 2020, which show patients suffering from four menstrual disorder namely Amenorrhea, Dysmenorrhea, Oligomenorrhea and Abnormal uterine bleeding. The Results show that in married females, ratio is 10% amenorrhea, 10% premenstrual syndrome, 60% Dysmenorrhea 20% Menorrhagea. In unmarried the ratio is 10% Dysmenorrhea, 60% amenorrhea, 20% Menorrhagia, 10% Metrorrhagia.

# CORONARY ARTERY BY-PASS/ANGIOPLASTY IS NOT A SOLUTION TO PROBE LIVER PHYSIOLOGY

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Angioplasty/Coronary Artery By-Pass is common practice in our prevailing medical system for protection of life from the life threatening coronary artery blockage (atherosclerosis, coronary artery disease or coronary heart disease) or lumen contraction leading to cardiac failure or heart attack. The formation of vascular thrombus or blood clot (which usually occurs) makes situation more drastic. The vascular thrombus grows under the endothelium of blood vessel, in its first step, by the Weakening of Endothelium under the stress of POISONOUS substances present in the blood stream. This weak endothelium gives way for the penetration or entry of Macrophages within the wall of vessel below the endothelium. Now the low-density lipo-proteins move to the interior of blood vessel from this weak endothelium and penetrate into these lodged cells, and along with it, cholesterol also settles down in the vicinity of these lodged debris. This plaque gradually grows and squeezes the lumen of the vessel. This is the partial story of the problem as several physically hard-working people taking complete protective medicines along with possible balanced food may get heart attack. A more frustrating situation is observed in those persons who have had coronary artery by-pass recently may also get growing arterial plaque and even contract heart attack. What is obscure as to how formation of this plaque growth is not seen in extremely carefree persons. I can quote two examples of heart attack after coronary artery by-pass that came under my treatment (1) a fellow of 49 year contracted severe category of heart attack just after 50 days after coronary artery by-pass and he was managed accordingly under homeopathic medication. (2) A young man of 34, he had had coronary artery by-pass 7 months after that he suddenly fell unconscious and the expert cardiologists recommended 2<sup>nd</sup> by-pass. They preferred homeopathic treatment at my clinic. These patients require an extremely wise, cautious and comprehensive treatment. For immediate protection of the heart and minimizing necrosis, the extra Oxygen supply and blood-plasma in the vicinity of the meager cardiac cells. A few homeopathic medicine play miraculous role i. e. Carbo vege 30, Vanadium 30 and Arnica montana 30 are given. Several cardiac homeopathic medicines are essential according to the condition of the patient that include Crataegus oxyacantha Q, Cactus 30, Naja 30, Spigelia 30 and Adonis vernalis 30 and several other medicines. Lastly a few medicines that normalize LIVER-physiology are also provided. These medicines impart positive results at the spot. These positives are observed through sphygmomanometer and stethoscope. These medicines are Chelidonium majus Q, Myrica Q Cheonanthus Q, Cholestrinum Q and Lycopodium 30 and etc. Here all medicines play their part according to their function, the demand of medicines that normalize the physiology of liver indicate that blood toxicity is not properly managed. In the atherosclerosis, the physiology of liver is not considered properly which is the root cause of the problem. The emergency condition is preferably coped with in the current methodologies of treatment instead of uprooting the actual causes of the trouble. I believe that the actual causes of the trouble ought to be tackled contrary to the make-shift and emergent tactics to slumber the issue.

# INCREASED RISK OF INFERTILITY, MARITAL MALADJUSTMENT AND PSYCHOLOGICAL MORBIDITY IN PCOS PATIENTS OF SOUTHERN PUNJAB, PAKISTAN

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The clinical manifestations of polycystic ovarian syndrome (PCOS) have been linked with psychotic morbidity and infertility in the women of fertile age. Considering limited research data available about the psychological aspects in these patients, we conducted current study to assess psychotic distress and infertility in PCOS patients of southern Punjab, Pakistan. In this study 204 PCOS women and 150 controls were enrolled. Data of recruited patients was collected from different hospitals of South Punjab (Bakhtawar Amin Memorial hospital Multan, Nishtar hospital Multan, DHQ Muzaffargarh, THQ Jatoi, Muzaffargarh and Indus hospital Muzaffargarh). A standardized questionnaire was administered in order to collect the data after obtaining formal consent and ethical approval. For statistical analysis of data, SPSS version 20 was used. Findings revealed that psychological morbidity was most common among infertile PCOS patients. The prevalence of anxiety (61.8% vs. 18.7%), depression (56.9% vs.15.3%), sleep apnea (35.3% vs. 6.7%), eating disorders (18.1% vs. 1.3%), marital maladjustment (27.5% vs. 5.3%), poor quality of life (35.3% vs. 3.3%) and migraine (55.4% vs. 10.7%) was significantly high in PCOS women as compared to controls (p<0.001). Infertility was significantly linked to psychotic implications and PCOS female (p<0.001). So, infertile PCOS patients were more depressed and anxious as compared to fertile. Infertility, especially primary infertility represents a major risk factor for psychological implications in Pakistani PCOS women of southern Punjab. Psychological disturbance was found to be more prevalent in married females as compared to unmarried females.

# EFFECT OF FERULA JAESCHKEANA ON ESTRUS INDUCTION AND CONCEPTION RATE IN ANOESTROUS AND DELAYED PUBERTAL ACHAI DAIRY CATTLE

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Herbs are considered as the oldest remedy for treating different diseases throughout the world. *Ferula Jaeschkeana* is a herb, common for its use in human reproductive disorders as well as for its estrogenic activity in lab animals. Here we attempted to explore the reproductive biological impact of *Ferula Jaeschkeana* in anestrous and delayed pubertal reproductive disorders as these reproductive disorders are associated with heavy economic losses of the livestock industry. Experiments were carried out in the 24 animals of native cattle breed with three doses of *Ferula Jaeschkeana* (30, 50 and 70 grams) which were shade dried, powdered and were given to the animals in their routine feeds as a bolus in morning and evening along with it blood was collected before and 3 days after the feeding of this herbs for the determination of progesterone and estrogen profile. As result of the trial animals showed the signs of behavioral estrus (restlessness, rising of tail, bellowing and mounting) and physical changes like the swelling of vulva and mucus discharge in 54.16% treated animals comprising of 33.34% animals from the delayed pubertal heifers group whereas 75% animals were from Anoestrus multiparous animals group. Likewise 0.08% animals in the delayed pubertal heifers group and 50% animals from Anoestrus multiparous animals group became pregnant and were confirmed on day 90 post breeding. Furthermore, the current study demonstrated the significantly high level of Serum estradiol hormone in tandem with significantly low level of progesterone hormone recorded in 50 and 70 grams *Ferula Jaeschkeana* feeding groups in comparison with the 30 grams and control group. Thus it could be deduced from the current study that feeding

50 grams *Ferula Jaeschkeana* herb roots in anestrous and delayed puberty Achai cows might be associated with the serum high level of estradiol hormone and low level progesterone hormone and resulting in the enhancement of the conception rates in The Achai cattle. Consequently, this herb may be used in the treatment of anestrous, silent estrus and delayed puberty in Achai dairy cattle under the existing management system.

#### STUDY ON BIOCHEMICAL PROFILE OF THE ABORTIVE WOMEN IN DISTRACT PESHAWAR

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Human pregnancy is the series of developmental events. However theses events are often associated with the metabolic alteration in the normal physiology of the maternal circulation which in turn resulted in the abortion. The abortion is associated with an enormous health, financial, and social burden on women in the developing countries including Pakistan. The retrospective case control, cross-sectional study was carried out during current dissertation. For this purpose a total of 60 pregnant women with age 18-40 years were selected randomly through convenient sampling techniques. Standard procedure was adopted during collection of the specimen that included the collection of the maternal blood, the above mentioned biological specimen was collected from 30 abortive pregnant women and 30 normal pregnant women. The biochemical indices were executed using the commercially available kits. The kits were used as per manufacturer 'instructions in the laboratory. During current study, the serum biochemical feature in the abortive women in the comparison with the normal delivered women were appraised. These serum biochemical feature included protein profile such as total protein, albumin, globulin, bilirubin, urea, uric acid; energetic profile i.e., glucose, cholesterol, triglyceride; mineral profile i.e., Phosphorous, calcium and magenium; enzymatic profile, i.e., AST, LDH and hormonal profile, i.e., Estrogen, Progesterone. The finding of current study demonstrated significant deviation from the normal in theses biochemical indices. TP(7.1±0.10g/dL vs 6.5±0.10g/dL), Chol(245.9±6.5mg/dl vs 162.6±9.5mg/dl), Mg(0.59±0.7mg/dl vs 2.1±0.14mg/dl), AST(23.3±1.1mg/dl vs 4.2±0.6mg/dl), Ph(5.2±1.9mg/dl vs 4.1±0.8mg/dl) concentrations increases at (P<0.05), while estrogen (3938.8±122.3 Pg/ml vs 1744.7±158.4 Pg/ml), progesteron(166.6±15.1 Pg/ml vs 56.3±5.3 Pg/ml), ALT(14.0±1.1mg/dl vs 2.1±0.2mg/dl), ALP(169.1±10.3mg/dl vs 59.6±4.3mg/dl), Ca(6.89±0.2mg/dl Vs 9.1±0.1mg/dl), Uric Acid(4.5±0.4 mg/dl vs 2.9±0.2 mg/dl), TG(234.6±11.3 mg/dl vs 107.9±4.4 mg/dl),  $HDL(61.7\pm2.3 \text{ mg/dl vs } 35.9\pm1.1 \text{ mg/dl})$ ,  $Albumin(3.5\pm0.10\text{g/dL vs } 2.0\pm0.10\text{g/dL})$ ,  $Globulin(3.4\pm0.0.1\text{g/dL vs } 1.0\text{g/dL v$ 1.5±0.1g/dL) concentrations were observed significantly reduced at (P<0.01) in abortive women when the comparison was made with the normal deliversies. On the other hand, Glucose (73.7±0.8mg/dl vs 93.2±5.6 mg/dl), creatinine (0.5±0.02 mg/dl vs 1.1±0.1 mg/dl), urea (10.2±0.35mg/dl vs 18.2±2.0 mg/dl), Total bilrubin (0.5±0.04 mg/dl vs 1.9 ±0.3 mg/dl), LDH (289.1±11.7mg/dl vs 515.8±10.5mg/dl,p=0.002) concentrations were significantly higher in the abortive women when compared with normal at (P<0.01).In conclusion, the current study demonstrated considerable variation in blood/serum biochemical factors in abortive women when compared with women having normal deliveries. These results signify a continuous existence of each biochemical factors in the normal deliveries, deviation of either lowered or higher from the normal biochemical range might be associated with causation of abortion. These parameters will help in finding diseases and will maintain the health of mother and fetus.

# STUDY OF RELATIONSHIP OF POLYCYSTIC OVARIAN SYNDROME WITH INFERTILITY AMONG FEMALE PATIENTS AT LADY READING HOSPITAL PESHAWAR

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Polycystic ovarian syndrome (PCOS) is a heterogeneous endocrine disorder affecting about 5-20% females of child bearing age. PCOS is characterized by Hyperandrogenemia, Oligoamenohhrea, Substantial number of immature

follicles, inadequate LH:FSH ratio, hirsitism, fibroids etc. The target of the present study is to ascertain the relationship of PCOS with infertility. For this purpose 100 respondents were studied in Lady Reading Hospital Peshawar. Data was collected through well fabricated questionnaire. The result revealed that women with PCOS had higher percentages of hirsutism(92%), acne(88%), fibroids(60%), depression(63%), imbalanced hormone(85%), irregular menstrual cycle(85%), heavy bleeding(73%), and (68 %) of women were facing difficulty to conceive a baby. It was collected that hirsutism, acne, depression, fibroids, hormonal imbalance, irregular menstrual cycle, and infertility are positively associated with PCOS. It was concluded that PCOS is the leading cause of anovulatery infertility among the females of child bearing age.

#### PREVALENCE OF DIABETES (A CASE STUDY OF PESHAWAR)

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The aim of present study was to find the prevalence of diabetes. As a non-communicable disease diabetes mellitus (DM) has major public health importance and its occurrence is swiftly growing all over the world at alarming rate. The current study was conducted to find the diabetes prevalence at Peshawar region in both urban and rural areas. The population studied consists of 100 families, 60 from urban and 40 from rural area. The questionnaire was filled from a responsible individual of each family, and data was obtained regarding diabetic and non-diabetic members. Out of 616 individuals of 100 families 61 individuals were found to be diabetic and 555 were non diabetic. Percentage of male diabetic individuals was recorded to be 12% and the percentage of female diabetic individuals was 8.70%. In urban areas a total of 354 individuals were studied among 60 families, 141 males 213 female, among these 38 (10.73%) were found to be diabetic, 19(13.47%) male and 19(8.92%) were female and a total of 316 (89.26%) were found to be non-diabetic. Similarly a total of 262 individuals were studied among 40 families of rural area, 119 were male and 143 were female, among these 23 (8.77%) were found to be diabetic, 11 (9.24%) male and 12 (8.39%) female, and a total of 239 (91.22%) were non diabetic. The above percentage revealed that the prevalence of diabetes is higher in males than in females. On the whole the diabetes prevalence for Peshawar population was 10.42%. The higher prevalence of diabetes calls for immediate attention towards prevention and health promotion, programs designed to reduce the load of this disease. Preventive measures should be taken to avoid the serious consequences of this disease. It is recommended that every person should follow healthy eating plan, be physically active, take medicines and check blood glucose levels on regular basis.

# PREVALENCE OF OSTEOARTHRITIS CAUSED BY THE DEFICIENCY OF CORTICOSTEROID HORMONE IN MALE AND FEMALE

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An attempt was made to study the possible role of corticosteroid hormones deficiency causing osteoarthritis. In this content, 15 patients suffering from the osteoarthritis were examined at wazir x-ray and Liaquat medical Hospital Jamshoro. According to the information sought the majority cases were old aged women and malnutrition persons. It was also noted that patients who had low in take of milk and had imbalance level of corticosteroid hormones were suffering from osteoarthritis. It is concluded that (i) Corticosteroid deficiency is the root cause of osteoarthritis (ii) The low milk consumption also effects the osteoarthritis (iii) The tissue cushioning the joint is deteriorate by many factors such as physical trauma, low level of corticosteroid hormones in the body (iv) Obesity is one of symptoms of OA (v) Irregular menstrual cycle in the patient is noted.

# ANALYSIS OF PANCREATIC GLAND DYSFUNCTIONING IN HYPOTHYROID PATIENTS IN DISTRICT GUJRANWALA, PUNJAB, PAKISTAN

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Hypothyroidism, characterized by low levels of thyroid hormones and elevated levels of TSH, affects the functioning of pancreatic gland by altering the levels of insulin secretion from pancreatic  $\beta$ -cells due to decreased production of thyroid hormones. The present study was aimed to analyze the relationship between hypothyroidism and pancreatic gland dysfunctioning Blood samples were collected from control and hypothyroid patients in district Gujranwala, Punjab, Pakistan. Blood samples of sixty normal and sixty hypothyroid patients were taken. Information regarding age, gender, weight, height, B. P was collected from participants. Inclusion and exclusion criteria were designed. Hypothyroid patients with high levels of TSH had significantly (P<0.05) low insulin and HDL cholesterol levels. However, the levels of fasting glucose, HOMA-IR, triglycerides, HbA1C and LDL cholesterol were significantly (P<0.05) high in hypothyroid patients indicating the diabetic condition. The current study indicated the high prevalence of pancreatic disorder in hypothyroid patients and thus, provided the clinical manifestation of the diabetes disease in hypothyroid patients by its early diagnosis and treatment.

### 6. TOXICOLOGY

### THERAPEUTIC EFFECT OF 2-(THIOPHEN-2-YL) 2,3 DIHYDROBENZOTHIAZOLE ON ROTENONE INDUCED PARKINSON'S RAT MODEL

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Rotenone is a pesticide and mitochondrial complex I inhibitor. It produces toxicity in humans and also in animals. The present study evaluated the therapeutic effect of 2-(thiophen-2-yl) 2,3 dihydrobenzothiazole against rotenone-induced Parkinson's disease. Rotenone was injected intraperitoneally at the dose of 1.5 mg/kg for 8 days. Administration of 2-(thiophen-2-yl) 2,3 dihydrobenzothiazole (10 mg/kg/day) was started before 15 days of rotenone injection. The effects of pre-treatment of 2-(thiophen-2-yl) 2,3 dihydrobenzothiazole were evaluated by different motor behavioral parameters (pole test, Kondziela's inverted screen test, inclined plane test, open field test, Rota rod test and footprint test). Determination of Hematological Parameters was also done by analysis of Blood and serum collections. Pre-treatment with 2-(thiophen-2-yl) 2,3 dihydrobenzothiazole reversed the gross motor impairments which were produced by rotenone. We conclude that 2-(thiophen-2-yl) 2,3 dihydrobenzothiazole, like its other candidate drug also protects against toxic effect of rotenone and can be beneficial against different neurodegenerative diseases.

# DETERMINATION OF LC50 OF MORINGA OLEIFERA ON KHAPRA BEETLE (TROGODERMA GRANARIUM E.)

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Trogoderma granarium (Everts), is known as world's most damaging pests of stored grain products (specifically wheat). Deltamethrin and phosphine are used in Pakistan to control its progeny, but these insecticides have lost their efficacy as this pest has developed resistance against them. Moreover, these insecticides have hazardous influence on human health. The aim of the present investigation was to find out the most effectiveness of lethal concentration of Moringa Oleifera leaf extract and exposure time period capable of controlling larvae infestation of T. granarium. The LC<sub>50</sub> value of 4<sup>th</sup> and 5<sup>th</sup> instar of Gujranwala population (obtained from lab culture) was calculated through mortality data and it was found that 4<sup>th</sup> instar larvae were the most resistant (showed highest LC<sub>50</sub>) while 5<sup>th</sup> instar larvae came forward as sensitive population (lowest LC<sub>50</sub>). Both instars were then proceeded to carry out biochemical analysis. The toxicity of Moringa leaf extract was determined on the energy reserves i.e, soluble protein, total protein contents and glucose contents. The obtained result showed the decline in total protein contents of 4<sup>th</sup> and 5<sup>th</sup> instar larvae at LC<sub>50</sub>. However, soluble protein contents and glucose contents were found to be elevated in both 4<sup>th</sup> and 5<sup>th</sup> instar larvae. Statistical analysis was performed to check the toxicity of Moringa leaf extract on T. granarium. Results obtained from this study showed that Moringa leaf extract had adverse effects on the energy reserves of T. granarium. This extract may be used to control the stored grain pests.

# HISTOPATHOLOGICAL CHANGES IN LIVER AND KIDNEY OF FRESHWATER FISH (CHANNA PUNCTATUS) INDUCED BY PERMETHRIN

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Permethrin is a broad spectrum synthetic pyrethroid insecticide recommended with restricted use due to its high toxicity for aquatic organisms. But increased use of permethrin in agricultural sector has resulted in increased insecticidal stress to freshwater fish. Thereby, current study was designed to investigate the histopathological changes of permethrin in liver and kidney of freshwater fish, Channa punctatus. Total twenty-five spotted snakehead fish (Channa punctatus) with 7-10cm length were divided into five groups (E0, E1, E2, E3 and E4) having five fish in each. Fish in group E0 were kept as control in uncontaminated fresh water, while those in group E1, E2, E3 and E4 were exposed to 0.25ppb, 0.50ppb, 0.75ppb, 1.00ppb of permethrin in fresh water, respectively for 96 hours. Water temperature observed during trial was 20-23°C, pH: 7.6-7.8 and dissolved oxygen was 100% saturated. Water and permethrin solution were renewed daily throughout experiment. At the end of experimental period, the morphometric parameters including total weight, total length, standard length and organ weight showed significant difference. Morphometric parameters have shown decrease by increasing permethrin toxicity. Samples from liver and kidney were histologically analyzed using microtomy. Hepatocytes showed degenerative changes including vacuolation, pyknosis, congestion of blood vessels, necrosis and infiltration of lymphocytes. While intestinal damage was observed as atrophy, blood congestion, detachment of villi, fusion of villi, goblet cell formation, shortening of villi and pyknosis. It was concluded that use of permethrin caused extreme toxicity to Channa punctatus so proper monitoring and care is recommended for its usage.

#### STUDIES ON ACUTE TOXICITY OF SILVER NANOPARTICLES TO LABEO ROHITA

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The effect of silver nanoparticles on histology and hematology of fish was evaluated. Seven healthy individuals of Labeo rohita were randomly distributed in each aquarium. Three different doses of silver nanoparticles were designated for three replicates i.e. 5 mg/l, 10 mg/l and 20 mg/l. Histological studies in organs like liver, gills, skin, kidney, and muscles were made to assess tissue damage due to silver nanoparticles after 96 hours of exposure. In gills fusion of primary and secondary lamella were observed. In muscles abnormal arrangement of muscle bundles was observed. In liver separation of macrophages and in kidney congestion of cells were observed. In case of skin inflammation of epidermis was observed. Biochemical parameters like serum protein, albumin, globulin and glucose were compared with normal control. In hematological parameters WBC, RBC, Hb, PLT, MCH and MCHC etc were also compared with normal control. Our research concluded that silver nanoparticles adversely affect the fish fauna. Physicochemical parameters of water were also checked during each trial.

# ZNO NANOPARTICLES INDUCE SEVERE LIVER DAMAGE AND MILD KIDNEY PROBLEM IN A MAMMALIAN MODEL

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Nanoparticles are most of the times intentionally synthesized particles ranging in size from 10-100 nm. Their relatively small size to large surface area make them as good loading and accessible tool in biomedicine. However, use of nanoparticles in modern biomedicine has faced a great deal of problems ranging from toxicity to alteration of genetic makeup. ZnO nanoparticles (ZnO-NPs) are the most widely used nanoparticles. Safe use of ZnO-NPs still needs careful and detailed investigations for their toxicity profiling. In the present study, toxic effects of orally administered ZnO-NPs were evaluated in albino mice for a period of 21 days. Histopathology (H & E), clinical biochemistry and bio-distribution analyses were performed after prerequisite period of treatment. ZnO-NPs at dose value of 250 and 500 mg/kg of the body weight induced toxicity in liver and kidney tissues. Inflammation and degeneration in liver hepatocytes was more pronounced than in kidney tissues. Also, few renal corpuscles, proximal and distal tubules were disturbed and enlarged. An increase in levels of ALT, ALP, AST, cholesterol and urea was observed, whereas a decrease in total proteins and albumin levels was recorded in the treatment groups. Zn content was increased in liver and kidneys, while there was no change recorded for the heart tissues.

# DETERMINATION OF LEAD CONCENTRATION IN SELECTED TISSUES OF *EUPHLYCTIS CYANOPHLYCTIS* (AMPHIBIA; DICROGLOSSIDAE) IN THATTA DISTRICT

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This study, determine the concentration of Pb (Lead) in kidney, liver and skin of adult *Euphlyctis cyanophlyctis* (Amphibia. The samples were collected from an urban area, Thatta city and an agricultural area, Mirpur Sakro. The water samples from the study areas were also collected. Samples were collected regularly during four seasons; Northeast monsoon, Pre monsoon, Southwest monsoon and Post monsoon. In Northeast monsoon, the contamination of Pb was at highest in liver of the individuals from Mirpur Sakro. However, the analysis of water samples showed low amount of Pb contamination. In pre monsoon, highest concentration of Pb was observed in liver, followed by kidney of the individuals from Thatta city while the lowest concentration was observed in the skin of the individuals from Mirpur Sakro. In South west and post monsoon seasons, highest concentration of Pb was noticed in the water samples from both study areas and both seasons. However, the concentration of Pb was relatively low in the tissues. The study reveals that the anuran fauna inhibiting, Thatta district are vulnerable to heavy metal toxicity in their environments. The outcome of this research will further provide the information about the conservation of frog species, and their role as the bio-indicators of respective environment.

### 7. VIROLOGY

### SURVEILLANCE OF HONEY BEE (APIS MELLIFERA) AGAINST VIRAL DISEASES IN DISTRICT KARAK

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Honey bees are characterized by their ability to produce honey that is a source of energy and have various medicinal benefits. Honey bee belongs to the family Apidae, in class Insecta. The present study aimed to find out "the surveillance of honey bee against viral diseases in district Karak Khyber Pakhtunkhwa". Twenty five apiaries (colonies =100%) were randomly selected in three main areas; Tehsil Karak, Takht-e-nasrati and Banda daud shah of district karak. Viral infected colonies were (1-19.3%) and non-infected colonies were (1-81.7%). They were further divided as managed apiaries (colonies=60%) and non-managed apiaries (colonies=39%). In managed apiaries infected colonies were separately arranged and regularly checked by beekeeper to minimize the transmission rate of viral infection. While the non-managed apiaries with viral infected colonies remain unchecked as beekeeper were unaware of the viral infection transmission. It was observed that the rate of transmission of infection is less in managed apiaries as compared to non-managed apiaries. Similarly, the survival of honey bees and the production of honey from managed aperies is higher than non-managed apiaries.

# DETECTION OF SARS-COV-2 IN CLINICAL AND ENVIRONMENTAL SAMPLES FROM LAHORE, PAKISTAN

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The surveillance of sewage water has become an extremely essential tool to trace the circulation of viruses in a population and to predict the outbreak of viral diseases. Sewage monitoring is more important especially for those viruses which cause subclinical infections since it is difficult to determine their prevalence. The present study aimed at investigating the presence of Novel coronavirus SARS-CoV-2, causing coronavirus disease (COVID-19), in sewage water collected from six drains of Lahore, Pakistan. Considering the high transmissibility and rapid spread of SARS-CoV-2, it was also planned to develop a robust and cheaper colorimetric RT-LAMP assay for naked-eye detection of SARS-COV-2 in sewage water. The viruses in sewage water were concentrated by PEG method before isolating viral nucleic acids. SARS-COV-2 was detected by RT-PCR. This data showed the presence of SARS-CoV-2 in all selected drains of Lahore. Furthermore, developed RdRp-based LAMP assay was successfully detect SARS-CoV-2 in clinical as well as sewage samples within 20 minutes. SARS-CoV-2 RNA was detected using our optimized LAMP assay in all of the sewage water samples. Likewise, LAMP assay described here could successfully detect the virus RNA in 26/28 (93%) of RT-PCR tested positive COVID-19 clinical samples with 100% specificity (n = 7) showing comparable efficiency to the qRT-PCR. The effect of various additives also tested on the performance of LAMP assay and found that addition of 10 mg/ml bovine serum albumin (BSA) could increase the sensitivity of assay up to 101 copies of target sequence. In conclusion, the present study is the first ever to describe the prevalence of SARS-CoV-2 in sewage water in the city of Lahore. Further, LAMP-based detection of SARS-CoV-2 in sewage as well as clinical samples could provide a sensitive first tier strategy for SARS-CoV-2 screening and can potentially help diagnostic laboratories in better handling of high sample turnout during pandemic situation.

# ALANTOLACTONE: A POTENTIAL MULTITARGET DRUG CANDIDATE FOR PREVENTION OF SARS-COV-2 CELL ENTRY

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The rapidly growing coronavirus throughout the world is associated with high death rates and transmission power. SARS-CoV-2 is a severe respiratory syndrome caused by a positive sSRNA enveloped virus. It was first diagnosed in Wuhan, China in December 2021. A total of 117 million cases were reported with 2.6 million deaths till March, 2021. SARS-CoV-2 has badly influenced the social and economy behaviors and created anxiety all over the world. Repurposing drug is a move toward to discovering the drugs with less time consumption and in a cost effective manner. The current investigation, proposed naturally occurring Alantolactone, member of sesquiterpene family as a potential SARS-CoV-2 inhibitor and it also inhibit various human protein involved in its entry into the cell and its biogenesis using molecular docking. Alantolactone efficiently binds with SARS-CoV-2 proteins including spike glycoprotein (S-protein), nucleocapsid protein (N-protein), main-protease and papain like proteases (pLpro), with binding affinity of -7.3, -7.9, -6.8, -7.1 kcal/mol, respectively. Alantolactone binds strongly to human receptor that is angiotensin converting enzyme-2 (ACE-2), S1-RBD & ACE2 interphase, Furin, AAK1, GAK and both closed and open configuration of TPC2 channel with binding energies of -6.7, -6.9, -8., -7.3 and -7.9 kcal/mol, respectively. Alantolactone binds with virus proteins and human targeted protein through hydrogen bonding and hydrophobic interactions. Alantolactone binds successfully with target viral proteins such that it binds with S-protein through hydrogen bonding with ARG1014 and through hydrophobic interactions with ALA766 and LEU1012 amino acid residue. It binds with N-protein through hydrogen bonding with GLN71 and hydrophobic interaction with PRO163 and LEU162 amino acid residues while it binds with Mpro through hydrophobic interactions with MET49, CYS145, MET165 and HIS41 amino acid residues. Alantolactone bound successfully to the human receptor protein ACE2 and makes hydrophobic interactions with PHE40 and PHE390 amino acid residues while it makes hydrogen bonding at amino-acid residue SER44 and hydrophobic interactions with TRP349 of S1-RBD complex. Alantolactone shows strong association with Furin by making hydrogen bonds with ASN310, GLN488 and GLY307 and through hydrophobic interactions with TRP531, PRO266 and TRP531 amino acid residues. Alantolactone tethered with GAK making hydrogen bonds with ASP27, PHE28, VAL29 and hydrophobic interactions with VAL45 amino acid residue. Alantolactone binds strongly with both open and closed states of TPC2. In open state configuration of TPC2, Alantolactone merges with binding region by making hydrophobic interaction with VAL294, PHE230 and PHE233 amino acid residues. However, Alantolactone forms hydrophobic interactions with PHE230, LEU229 and LEU556 amino acid residues. Molecular docking simulations and the ADMET properties and toxicity predictions suggest that Alantolactone could effectively binds with various viral target protein as well as human target proteins and could be developed into a novel SARS-coV-2 inhibitor. However, the further in-vivo & in-vitro studies are mandatory to confirm Alantolactone against SARS-coV-2.

### INCIDENCE OF COVID-19 AMONG THE POPULATION OF DISTRICT ATTOCK

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Corona virus is another pandemic with almost no data accessible about its transmission and relationship with ecological factors. In comparison with known corona viruses, SARS-CoV-2 that can affect individuals is defined as the seventh corona virus with certain structural differences. The episode of COVID started as the unknown reason of pneumonia in Wuhan, China, in December 2019, which has been presently scattering quickly to other countries out of Wuhan. Objective of the recent research was to determine the incidence rate of Corona virus among the population of Attock city, Punjab.This retrospective hospital-based observational study used data from 01 April 2020 to 07 December 2020. A sum of 22,962 supposed individuals for Corona virus visited Government hospitals and laboratories in District Attock, Pakistan. Pharyngeal and nasal swab specimens were taken and tested for viral nucleic

acid by using real-time reverse-transcriptase polymerase chain reaction assay (RT-qPCR). The total positive confirmed cases out of 22,962 suspected individuals were 843(3.67%). Out of those confirmed positive cases, 367(43.53%) were reported from Tehsil Attock, 94(11.15%) Were from Tehsil Fateh Jang, 51(6.05%) were from Tehsil Hassanabdal, 211(25.03%) were from Tehsil Hazro, 30 (3.56%) were from Tehsil Jand 59(7%) were from PindiGheb, and 31(3.68%) were from Travelers. The overall number of deaths is 22 (2.61%), active cases were 96 (11.39%), and recovered patients were 725 (86%). It was concluded that the incidence rate is maximum in Thesil Attock and minimum in Thesil Jund as compared to other Thesil of District Attock.

### 8. FORENSIC SCIENCES

### INSECT SUCCESSION AND DECOMPOSITION PATTERN ON ORYCTOLAGUS CUNICULUS CARCASS

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Insect succession and decomposition pattern can be used as a tool for the calculation of Post Mortem Interval Analysis (PMI) in medico-legal issues. The present study was conducted to determine the insect succession pattern and decomposition stages on domesticated rabbit (*Oryctolagus cuniculus*) carcass at Dir, Pakistan. Five decomposition stages were detected which were fresh, bloated, active decay, advanced decay and dry remains stage. A total of 8 different insect species were recorded on the rabbit carcass. The most common and abundant insect species that visited the carcass was *Chrysomya megacephala* (Calliphoridae). Temperature and relative humidity in the study area were directly correlated with the insect succession and decomposition pattern on the rabbit carcass. Findings of the present study can be used for the calculation of Post Mortem Interval in resolving medico-legal issues and for crimes scene detection in forensic analysis. This study can be conducted on different animal models that resemble more to human beings in order to get more precise results for the calculation of PMI. Further studies are recommended to cover a wide spectrum of different ecological and topographical areas with different climatic conditions for obtaining a more precise model to resolve medico-legal issues with the help of forensically important insect fauna.

### 9. OTHERS (LATECOMERS)

#### BIOCONTROL POTENTIAL OF CRY 1C GENE AGAINST LEPIDOPTERAN PESTS

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The prospective about Bacillus thuringiensis in managing different insect pests, especially Lepidoptera has been reported. Bacillus thuringiensis-based product having old background shown its nontoxic character and applications in agrochemical industries. A number of Bacillus thuringiensis insecticides have been reported for field applications. Various crystal proteins are obtained from Bacillus thuringiensis. These crystal proteins have unique insecticidal activity and are pathogenic to many insect orders, such as Lepidoptera, Diptera, Coleoptera etc. Numerous Bacillus thuringiensis strains have been reported. A unique crystal protein is obtained by each Bacillus thuringiensis strain. Bacillus thuringiensis crystal proteins encoded by various transgenic plants. Toxins formed by Bacillus thuringiensis have been examined. These Bt. toxins have high toxicity against harmful insect pests. The toxicity of Bt. toxins against Lepidoptera pest reported. These insecticidal proteins are active against many insect pests. These insect pests are harmful for the development and final yield of Bt. crops. Bacillus thuringiensis isolates have high resistance against Lepidoptera pests. So, Bacillus thuringiensis insecticides have successful application in the management of Lepidoptera pests. The lethal concentrations (LC values) of Bacillus thuringiensis strains have been calculated. Bacillus thuringiensis proteins have historical importance with broad range of agrochemical and many other applications. They may be used for drug or gene delivery, environmental protection and detection of human fatal diseases, such as cancer etc.

### EVALUATION OF CYTO-PHYSIOLOGICAL EFFECTS INDUCED BY METHOMYL IN CTENOPHARYNGODON IDELLA

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It is well known fact that pesticides are important tools of modern agriculture used to shield crops from pests and in public health to prevent diseases. These pesticides enter the aquatic ecosystem in whole or in residual form, posing a major threat to aquatic organisms in general and fish in particular. In this connection the present investigation was undertaken to evaluate the cyto-physiological alterations induced by methomyl (insecticide) in Ctenopharyngodon idella. Fish were divided into four groups. One group was considered as control group and was kept without treatment while the other three groups were treated with different concentrations (0.2 mgL-1, 0.8 mgL-1 and 1.2 mgL-1) and time duration (3,6,9,12,15 days) against methomyl respectively, and observed the significant modulations in haematological, histopathological features and red blood cells and nuclear anomalies in fresh water fish (Ctenopharyngodon idella). The red blood cells count (RBC), hemoglobin (Hb), hematocrit (Hct), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC), neutrophils, eosinophils and monocytes concentrations were declined in the fish exposed to various concentrations of methomyl as compared to control group. However, mean corpuscular volume (MCV), total leucocyte count, lymphocytes, platelets were observed to be significantly higher. Histopathological alterations in the gills were epithelial lifting, curling, erosion of secondary lamellae, blood congestion in the secondary lamellae, fusion of secondary gills lamellae and disruption of cartilage core. In the liver histopathological alterations were lymphocyte infiltration, cloudy swelling of hepatocytes, degeneration of hepatocytes, necrosis in hepatpcytes, cytoplasmic vacuolation and blood congestion. Brain tissue reported anomalies as blood congestion, degeneration of neuronal cells, leukocytic infiltration, necrosis and vacuolization. In the intestine histopathological alterations were hemorrhages, leukocytic infiltration, necrosis, fusion of villi, detachment of villi and villi sloughing. Similarly in kidney tissue abnormalities were expansion of space inside the bowman's capsule, degeneration of bowman's capsule, necrotic changes, cytoplasmic vacuolization and glomerular shrinkage. While in muscle tissues histopathological alterations were degeneration in muscle bundles, splitting of muscle fibers, vacuolar degeneration, muscle oedema, zig-zag of muscle fibers, necrosis of muscle fibers and lesion in muscle tissues. Various red blood cells and nucleus anomalies including deformed cell, swollen cell, hemolyzed cell, deformed nucleus, nuclear shift, and micronucleus were observed against different concentrations of methomyl. It is obvious that the various concentrations of methomyl caused significant variations in hematological indices and irreversible changes in histology of gills, liver, brain, intestine, kidneys and muscle of fish, while various red blood cells and nucleus anomalies were also reported that indicate the DNA damage in the nucleus of red blood cells.

# IMPACT OF SELECTED PARAMETERS EXAMINING THROUGH HEMATOLOGICAL SAMPLES IN LOCAL POPULATION OF RAZZAR, SWABI

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Specific hematological references are used for specific population ad used for diagnosis and treatment purposes. According to age wise study the Reference interval of male show significant difference with RBCs, HGB, MCHC, Lymphocytes% and P-LCR while female R1 show only significant difference with P-LCR. Male RBC reference value at age group 15-25 were 4.63×1012/L (2.35-5.90×1012/L), 26-35 were 4.59×1012/L (4.43-5.89×1012/L), 36-45 were  $4.60 \times 1012/L$  (3.69-5.52×1012/L), 46-55 were  $4.49 \times 1012/L$  (2.35-6.72), 56-65 were 4.271012/L (2.38-5.25×1012/L), with P-value (0.005). HBG at age 15-25 were mean values 12.16g/dL (6.40-15.90g/dL), at age 26-3511.92g/dL (6.60-16.10), at age 36-45 were 11.89g/dL (5.90-16.10g/dL), at age 46-55 were 12.09g/dL (5.90-15.90g/dL), at age 56-65 were mean value 10.80g/dL (5.80-14.80g/dL) with a p-value (0.011). MCHC at age 15-25 were 33.10g/dL (28.60-72.40g/dL), 26-35 were 32.10g/dL (26.90-37.80g/dL), 36-45 were 32.07g/dL (24.60-37.50g/dL), 46-55 mean value 33.38g/dL (22.10-39.30g/dL), 56-65 were 31.64g/dL (27.50-33.90g/dL) with p-value (0.039). Lymphocytes at age 15-25 mean values were 26.35% (6.80-42.50%), 26-35 were 24.8% (9.30-49.50%), 36-45 were 27.63% (8.20-53.10%), 46-55 mean value 31.98% (9.70-81.00%), 56-65 were 28.69% (5.80-64.50) with Pvalue (0.029). P-LCR at age 15-25 mean values were 32.15% (16.70-46.10%), at age 26-35 were 29.06% (2.40-49.50%), at age 36-45 were 31.08% (14-62.50%), at age 46-55 were 33.64% (18.50-67.60%) and 56-65 were 29.06% (13.60-49.20%) with P value 0.042. Age wise reference value of females, the P-LCR only show statistically significant difference with respect to blood parameters having age groups 15-25 P-LCR mean value 30.75% with minimum and maximum value (16.60-60.30%), at age 26-35 having 28.36% (18.50-50.20%), at age 36-45 having value 30.51% (15.20-53.20%), at age 46-55 having mean value 29.10% (16.70-45.70%), and at age 56-65 mean value 33.48% (15.60-51.90%) and show statistically significant relation with blood parameter P-LCR having P-value (0.040). The Reference 1nterval for both male and female according to gender wise study include 250 males and 250 females with minimum and maximum limit. For male the RBC 4.51×1012/L (2.35-6.72×1012/L), for female RBCs 4.32×1012/L (2.46-6.16×1012/L) having P-value (0.000). HBG for male 11.77g/dL (5.80-16.10%), HGB for female 11.12g/dL (4.12-15.30g/dL) having P-value (0.000). HCT for male 36.20% (3.93-55.90%), HCT for female 34.70% (11.40-48.60%) having P-value (0.003). MCHC for male 32.46g/dL (22.10-72.40g/dL), MCHC for female 31.93g/dL (24.70-37.20g/dL) having P-value (0.032). Lymphocytes for male 27.90% (5.80-81.00%), Lymphocytes for female 26.05% (4.30-59.70%) having P-value (0.045). Monocytes for male 7.98% (3.10-20.40%), Monocytes for female 7.25% (2.70-22.20%) having P-value (0.002). Neutrophil for male 63.95% (12.20-87.40%), Neutrophil for female 66.38% (4.40-87.60%) having P-value (0.011). These parameters show significant difference with respect to gender wise study. Total 500 samples included 87 smokers and 413 non-smokers in which the White blood cells having Mean value for smokers is 10.18×109/L (3.20-40.20×109/L), for non-smokers mean value of WBCs 9.19×109/L (1.30-35.30×109/L) with P-value (0.043) and Monocytes having Mean value for smokers 8.18×109/L (3.10-20.40×109/L), for non-smokers Monocytes Mean value 7.49×109/L (2.70-22.20×109/L) with P-value (0.029) show statistically significant association with blood parameters. HBG and MCHC show statistically significant relation with marital status. Total 500 samples having 358 married while 142 unmarried. The Hemoglobin (HBG) mean value of married 11.30g/dL having minimum and maximum value (4.12-16.10g/dL), while unmarried having mean value

11.83g/dL (4.30-16.10g/dL) with P-value (0.010). Mean corpuscular hemoglobin concentration (MCHC) also show significant association with blood parameters having mean value of married are 32.02g/dL (22.10-38.10g/dL), while unmarried having mean value 11.83g/dL (24.70-72.40g/dL) with P-value (0.021). Some hematological parameters of blood show statistically significant relation with sleeping duration of individuals. These parameters were Hematocrit (HCT), Red Distribution Width-CV (RDW-CV), White Blood Cells (WBC), Monocytes (MID), and Lymphocytes (LYMP). The Mean value of Hematocrit at 5 hours sleeping duration is 32.52% with minimum and maximum values (3.93-47.50%), at 6 hours sleeping duration 36.52% (24.10-55.90%), at 7 hours sleeping duration mean value 35.26% (16.40-45.60%), at 8 hours sleeping duration 35.93% (18.20-48.60%), at 9 hours sleeping duration 35.01% (19.30-47.20%), and more than 9 hours sleeping duration Mean value 36.23% (19.90-44.90%) having P-value (0.025). The Mean value for RDW-CV at 5 hours sleeping duration is 14.85% (11.90-35.30%), at 6 hours sleeping duration 13.38% (11.10-19.40%), at 7 hours sleeping duration 13.62% (11.50-23.10%), at 8 hours sleeping duration 13.47% (11.80-17.5%), at 9 hours sleeping duration 13.70% (12-20.50%), at more than 9 hours sleeping duration mean value is 13.50% (12-19.80%) having P-value (0.004). The Mean value for White blood cell at 5 hours sleeping duration is 11.43×109/L (6.40-35.30×109/L), at 6 hours sleeping time 8.31×109/L (1.50-18.90×109/L), at 7 hours sleeping duration mean value 9.48×109/L (2.10-40.20×109/L), at 8 hours sleeping duration 9.32×109/L (1.30-31.30×109/L), at 9 hours mean value 9.15×109/L (3.20-28.90×109/L), at more than 9 hours sleeping time the mean value for WBCs 9.63×109/L (2.90-22.90×109/L) having P-value (0.016). The Mean value for Monocytes at 5 hours sleeping duration 7.94% (4.40-46%), at 6 hours sleeping duration 6.82% (3.10-64.5%), at 7 hours sleeping duration 7.93% (2.70-81%), at 8 hours sleeping duration 7.50% (3.20-57.70%), at 9 hours sleeping duration 7.51% (2.80-59.70%), at more than 9 hours sleeping duration the mean value of monocytes 8.24% (3.90-65.90%) having P-value (0.041). The Mean value of absolute Lymphocytes at 5 hours sleeping duration is 5.02×109/L (1.20-81.70×109/L), at 6 hours sleeping duration 2.28×109/L (0.30-9.20×109/L), at 7 hours sleeping duration 2.43×109/L (0.60-8.00×109/L), at 8 hours sleeping duration 2.26×109/L (0.30-9.20×109/L), at 9 hours sleeping duration 2.40×109/L (070-7.50×109/L), at more than 9 hours sleeping time the mean value for Lymphocytes is 2.55×109/L (0.890-12×109/L) having P-value (0.005). These values show statistically significant difference with hematological blood parameters. The socio-economic status and exercise show difference within groups but show no statistically significant differences.

### TOXICITY OF BACILLUS THURINGIENSIS AND CRY2AB GENE AGAINST LEPIDOPTERAN PESTS

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Vegetables are an essential source of nutrients and commonly used as a food regularly. The yielding ratio of crops are affected by many lepidopteron species act as a pest. The present study were designed to analyze the biocidal activities of endotoxins produced by Bacillus thuringiensisfrom cry2Ab gene against commonly found lepidopteron pests (Spodoptera litura, Helicoverpa armigera, and Chilo partellus). The vegetable plants were grown in a confined field trails under insect proof netting. Similarly, the Bacillus thuringiensis(Kurstaki species) having active cry2Ab were grown in control laboratory conditions using L.B media to get maximum colonies for formulation of spore/crystal mixture with varying concentration (100µg/ml to 1000 µg/ml). Also, different pest's larva were reared in control laboratory conditions. After experimentations, the calculated L.C50 (96-hours) of cry2Ab gene spore-crystal mixture against different lepidopteron pests were 138.95µg/mlfor H. armigera, 277.27µg/ml for S. litura, and 106.72µg/ml for C. partellus respectively. The comparative potencies analysis results revealed that the endotoxins mixtures of cry2Ab shows high potency of 100% mortality caused inC. partelluswith 400μg/ml followed by H. armigera with 350 μg/ml, and S. litura with 700μg/ml. The overall results indicated that the Bt. cry2Ab gene endotoxins provide an effective and sustainable results in order to prevent from economical loss of crops yielding through taking control of lepidopteron pests. Based on our research findings, the Bt. cry2Ab gene endotoxins biocidal activity against different lepidopteron pests shows promising outcomes for controlling of pests in pest managements.

# EFFECT OF PARAQUAT ON CYTOPHYSIOLOGICAL PARAMETERS OF GRASS CARP (CTENOPPHYRENGODON IDELLA)

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Pesticides are particularly used to reduce the number of pests and increase productivity of crops but intensive utilization of these chemicals has deleterious impact on non-targeted organisms. Pesticides drain directly or indirectly to aquatic bodies and greatly disturb ecosystem of aquatic fauna. Among aquatic organisms, fish are greatly affected by pesticides and it induce several physiological and biochemical alterations. As important component of food chain, human being also faces severe health problems by feeding on contaminated fish. The present study was designed to explore various histo-physiological anomalies of grass carp (Ctenohpharyngodon idella) against the various toxic sub-lethal concentrations of paraquat (herbicide). Grass carp was exposed to 0.02, 0.04 and 0.06 mg/L paraquat for 04, 08 and 06 days respectively. The present study unveiled the toxic impact of paraquat by exploring the decrementing level of hemoglobin, hematocrit, mean cell hemoglobin, mean corpuscular volume and red blood cells while increase in concentration of leucocytes and platelets were observed. Besides these alteration red blood cells and nucleus abnormalities were also observed including swollen nucleus, terminal nucleus, damaged red blood cells and micronucleus. Similarly, various tissues abnormalities were observed as; in gills tissues cellular necrosis, irregular cells, inflammatory cell and degeneration of epithelial cells were observed, while swelling of laminar propria, large lumen and flattened villi were observed in intestine tissues. Other notable histological alterations in kidneys were hemorrhage, enlarged bowman capsule while pyknosis were observed in liver tissues. From our study it has been concluded that herbicide find their way to reach the aquatic body where it alters the ecosystem and cause severe damages to aquatic fauna. Beside these it also effects human health when they feed on contaminated fish. Therefore, all pesticides should be used in a control way in order to prevent aquatic pollution.

# THE ADVERSE EFFECTS OF MALATHION ON HISTO-HEMATOLOGICAL PARAMETERS OF GRASS CARP (CTENOPHARYNGODON IDELLA)

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Pesticides are widely utilized against pests to reduce the deleterious impact of pest and increase productivity of crops. Excessive uses of these chemicals ultimately become harmful to other organisms including human beings. Fish as the occupant of the aquatic bodies cannot stay away from the harmful impact of these chemicals. Municipal Solid Waste (MSW) comprises solid waste from households, commercial area, construction waste and chemical waste that were not drown in open places. They are highly degradable and easily dissolve with air, which can disperse and contaminate water result in increased risk of pollution in aquatic bodies. In the present investigation, grass carp (Ctenopharyngodon idella) was exposed to various concentrations of malathion (insecticides), 0.02, 0.04 and 0.06 mg/L for 4, 8 and 12 days respectively. Results indicated that blood indices alteration is dose dependent like with increase in concentration of malathion, elevated level of leucocytes and platelets were observed while hemoglobin, hematocrit, mean cell hemoglobin, mean corpuscular volume and red blood cells level decreases. To cope with the stress environment, white blood cells (WBC) concentrations were incremented. Blood samples were also checked for red blood cells and nucleus abnormalities. Among red blood cells and nuclear anomalies infected cells, swollen nucleus and karyopikonisis were reported. Malathion induces severe histological disorders in grass carp including necrosis, inflammatory cells and swollen cells in gills while hemorrhage, degeneration of bowman capsule in kidney tissues of grass carp. Similarly, various histological alterations were reported in liver like larger vacuoles, pyknosis while large lumen and cellular necrosis were reported in intestine tissues.

### **SECTION – II**

### PESTS AND PEST CONTROL

### DIETARY HABITS OF SHORT TAILED MOLE RAT (NESOKIA INDICA) INHABITING AGRO-ECOSYSTEM OF POTHWAR PLATEAU, PAKISTAN

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The present study was designed to investigate the dietary habits of short tailed mole rat inhabiting croplands of Pothwar Plateau, during wheat and groundnut cropping and non-cropping season. The rodent specimens were captured from four selected sampling sites (one from each district) Rawalpindi, Attock, Jhelum, Chakwal of the Pothwar Plateau, the samples were collected from groundnut, and wheat fields and also from the follow lands inside the crop fields and on the field boundaries, by using both kill and live traps, baited with the pieces of guava, peanut butter, bread, tomato, chapatti with pickle. The stomach contents of short tailed mole rat were analyzed using micro histology. The plant species consumed were identified by preparing light microscopic slides of the obtained material and comparing it with that of reference material that was collected from sampling area. The specimens were captured from wheat, groundnut, and fodder crops and from fields' boundaries. The trapping results revealed that the success of capturing was higher on guava bait then the rest of others. Trapping success was higher in cropping season of wheat (n= 7), followed by peanut (n= 5). During non-cropping season the success were low and mostly from field boundaries. The results of the analysis of 18 stomachs contents of short tailed mole rat showed that the rodent species mostly feeds on cultivated crops (wheat, groundnut) during their cropping seasons. Simultaneously it also consumes some fodder crops like millet, barley and sorghum etc. The diet of the species mainly comprised of cultivated crops, along with varying proportion of wild vegetation. The wild vegetation species in the study area may serve as staple food of the short tailed mole rat, sustaining it throughout the year, especially during the non-cropping seasons of wheat and groundnut crops.

### DIETARY HABITS OF LESSER BANDICOOT RAT (BANDICOTA BENGALENSIS) IN AGRO-ECOSYSTEM OF POTHWAR PLATEAU

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The rodents are widely distributed and serious agricultural pests in Pakistan. In the current study we investigated the dietary habits of lesser bandicoot rat (*Bandicota bengalensis*) in agricultural crops of the Pothwar Plateau and its variation during cropping and non-cropping seasons. Four potential sampling sites (one from each district) were selected through reconnaissance survey for data collection. The research activities were conducted at major field crops including wheat and groundnut and in the fallow lands during the crop and the non-crop seasons both on the field boundaries and inside the crop fields. The trapping was carried out using kill traps baited with the pieces of fresh fruit like guava, tomato and also used chapatti with pickles and peanut butter to avoid bait shyness.

Diet composition of the rodent species was studies using stomach contents analysis, applying micro-histological technique. A total of 20 rodent specimens were trapped; the success was higher in cropping season of wheat (n= 8), followed by groundnut (n= 6). During non-cropping season, the success were low and the capturing was done on the field boundaries, four specimens were captured from the field boundaries of the wheat and two from the field boundaries of the groundnut. Stomach contents analysis of specimens revealed that the bandicoot rat predominantly consumed cultivated crops during cropping season whereas weeds were recovered from stomach samples of specimens captured during non-cropping season. The rodent species consumed both agricultural crops and wild vegetation. Among crops, it most frequently consumed wheat (*Triticum aestivum*; 28.57%), followed by groundnut (*Arachis hypogea*; 11.26%), sorghum (*Sorghum bicolor*; 10.17%), chickpea (*Cicer arietinum*; 9.52%), maize (*Zea mays*; 6.49%), millet (*Pennisetum glaucum*; 5.84%), barley (*Hordeum vulgare*; 4.98%), and mustard (*Brassica campestris*; 4.98%). While among wild vegetation it utilized khbal gha (*Cynodon dactylon*; 7.79%), baron dhab (*Demostachya bipinnata*; 7.36%) and Prickly flower (*Achyranthes aspera*; 3.03%). The study concludes that along with the wheat and groundnut crops, grasses, weeds, and some fodder crops are also an important component the diet of bandicoot rat.

### DIETARY HABITS OF INDIAN GERBIL (*Tatera indica*) INHABITING AGRO-ECOSYSTEM OF POTHWAR PLATEAU

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In the current study we investigated the dietary habits of Indian gerbil (Tatera indica) in agricultural crops of the Pothwar Plateau during cropping and non-cropping seasons. Four potential sampling sites (one from each district) were selected through reconnaissance survey for data collection. The research activities were conducted at major field crops including wheat and groundnut and in the fallow lands during the crop and the non-crop seasons both on the field boundaries and inside the crop fields. The rodent specimens were trapped using kill traps baited with the pieces of fresh fruit like guava, tomato and also chapatti with pickles and peanut butter to avoid bait shyness. The diet composition of the rodent species was studied through analysis of its stomach contents, applying micro-histological technique. A total of 30 specimens were trapped by using snap traps in wheat, groundnut and adjacent fodder crop of the study area. The reference light microscopic slides of the reference material (crops and wild vegetation) and those of stomach contents were prepared and compared. Results revealed that Indian gerbil rat is largely omnivore and fully dependent on the surrounding area with proportions of major dietary items (insects, herbs, shrubs, grasses and seeds) varied among crop stage and seasons. The diet of this rat species heavily depended on the wheat plants and grains for their food as the crop approaches maturity. During non-crop season the most frequently consumed food item by this rodent species was Ziziphus nummularia (19.89%), followed by Cynodon dactylon (16.02%), Sorghum bicolor (14.89%), Zea mays (13.57%), Brassica campestris (12.23%), Pennisetum glaucum (10.56%), Arachis hypogaea (10.03%), Hordeum vulgar (8.54 %), Solanum nigrum (8.67%), Achyranthes aspera (6.01%), Eruca sativa (5.78%), Medicago spp (5.67%), Desmostachya bipinnate (2.02%) and Artemisia dubia (0.89%). The summer diet (non-crop season) of this rodent was relatively less diversified than that of the winter diet (cropping season). Insects (animal matter/ crop pets) were consumed in large quantities highlighting the positive biological control of Indian gerbil in the agro-ecosystem of the Pothwar Plateau. The findings of the current study indicate that the Indian gerbil is a generalist consumer and it can change its feeding habits, depending upon the availability of food materials and can cause heavy losses to the crops and as a result the populations of the rat needs to be controlled through appropriate methods and techniques.

# EVALUATION OF SOME AQUEOUS PLANT EXTRACTS AGAINST PLANT FEEDING THRIPS (*THYSANOPTERA*) AND MITES (*ACARI*).

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Mites and thrips are most destructive pests of ornamental crops, fruits and vegetables. Many botanicals are recently considered to be effective against different sucking insect pests. In current experiment, effectiveness of aqueous solutions of Garlic (*Allium sativum*), Mint (*Mentha*), Onion (*Allium cepa*) and Marigold (*Calendula officinalis*) were evaluated against two spotted spider mites (*Tetranychus urticae*) and onion thrips (*Thrip tabaci*). Five treatments with three replications (CRD) against each pest were considered with different concentrations of each aqueous solution ( $T_1 = 10\%$ ,  $T_2 = 5\%$ ,  $T_3 = 2.5\%$  and  $T_4 = 1.25\%$ ) and ( $T_5$ ) as control. These prepared aqueous solutions were applied on leaves and used for making thrips and mites cells. The data was collected after 24 hours, 48 hours, 72 hours and 96 hours against pest populations. The tendency of mortality by given solutions were observed and it was for garlic (90.00) > onion (81.67)  $\geq$  marigold (81.67) > mint (73.33) respectively. Maximum mortality for each of the aqueous solution was obtained by using (10% aqueous extract) in  $T_1$  and least was obtained by using  $T_4$  (1.25% aqueous extracts). Further, the mortality percentages were more in case of *T. urticae* as compared to *T. tabaci*.

### IMPACT OF PEST MANAGEMENT PRACTICES ON INSECTS AND BIRDS ASSOCIATED WITH SUNFLOWER CROP

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Sunflower (*Helianthus annuus* L.), an important oilseed crop in Pakistan, is subjected to ravages of biological factors, such as insects and birds. Effect of insect and bird pest management vice versa has not been reported. Therefore, current study was planned to investigate the effect of insecticides, Proclaim (T1), Chlorpyrifos (T2) and Malathion (T3), and bird repellents, Reflecting ribbons (T4), T5 (reflecting mirrors T5) and scarecrow ( $T_6$ ) on insects and avian species, collectively. T7 was considered as control group. For this purpose, land of 3 kanals was selected at University of Agriculture, Faisalabad and was divided in 3 blocks of three varieties (A, B and C). Each block was further divided into 7 sub plots by using split block design of RCBD. Each treatment was applied in triplicate. Sunflower seeds were obtained from Ayub Agriculture Research Institute, Faisalabad and were tested by using seed flotation technique before sowing. The results were analyzed statistically by using mean values and ANOVA. Results showed the effectiveness of flotation technique and 90% - 95% seeds of each variety were viable. The results showed significant effect of T1, T2, T3, T4, T5 and T6 on insect's population reduction as compared to control group where the insect infestation increased continuously. Birds' attack on all three varieties of sunflower, was significantly reduced in all treatments, and treatments were helpful in repelling both insects and birds. Birds escaped direct mortality due to pesticides application. Economics of treatments has also been taken into consideration, and both can have significant effect.

### EVALUATION OF WOOD DRYING METHODS FOR SEASONING TO ENHANCE RESISTANCE AGAINST TERMITES

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In the present work, sap wood of *populous deltoids*, *Syzygium cumini*, and *Mangifera indica* were seasoned by oven drying, kiln drying and sun drying in order to minimize moisture contents less than 20% against its starting

moisture contents. In first experiment, the timber stakes of M. indica, S. cumini and P. deltoids of size 10cm×5cm×2cm were prepared. Seasoning were conducted under sun for 7-15 days, in oven at 100°C, 150°C and 200°C for 10-15 days and also in solar kiln for 10 and 15 days starting from March, 2018 onward. Also the physical and chemical characteristics of timber were tested which include the moisture content, density, cellulose and hemicellulose level of treated wood. Results of this trials showed that Syzygium cumini timber dried in oven have minimum weight loss which was 4.821% at 120 exposure days while *Populus deltoids* timber dried under sun showed maximum weight loss which was 75.500% at 60 exposure days. Conducted the second experiment to check the penetration of oils. We compared penetration neem and linseed oil between two different dipping time 36 and 72 hours. Results revealed that S. cumini seasoned wood have maximum oil penetration after 72 hours of dipping which was 77.524% at 120 exposed days while P. deltoids timber showed minimum penetration in 36 hours which was 14.062% after 60 days. A second comparison revealed that wooden stakes treated in oven drying showed maximum penetration of neem oil in 72 hours of dipping which was 77.460% after 120 exposed days while sun dried timber stakes have minimum linseed oil penetration in 36 hours of dipping time which was 13.062% after 60 exposure days. The third experiment was performed to find out penetration of insecticides in treated wooden stakes. We also made a comparison of imidacloprid and fipronil insecticides after two different application time 36 and 72 hours. Results from this experiment revealed that S. cumini timber have maximum insecticide penetration at 72 hours of application which was 76.3% after 120 exposure days while P. deltoids stakes showed minimum insecticide penetration at 36 hours of application which was 10.1% after 60 days. Another comparison confirmed that maximum penetration of fipronil insecticides observed in oven dried stakes which was 84.5% after 120 exposure days while sun dried wooden stakes showed minimum penetration of imidacloprid in 36 hours of application which was 7.9% after 60 days. The moisture contents of these stakes calculated in each treatment. Before putting seasoned woods in the underground pits, these weighed, labelled and replicated thrice for each treatment. Data on weight loss of wooden blocks in each experiment recorded after exposure to the termite at 60 and 120 days. In all these experiments treatments were arranged with Completely Randomized Design (CRD). The obtained data from all above experiments were analysed by using Minitab 17 software.

# THE LEVEL AND DISTRIBUTION OF SELECTED ORGANOCHLORINE PESTICIDES IN WATER OF RIVER SATLUJ PAKISTAN

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The study was aimed to identify different environmental factors (selected organochlorine pesticides) affecting the river water of Satluj. River Sutlej is about 1400 kilometers long and its water is extensively used for irrigation in Punjab, located both in India and Pakistan, which was also a reason of dispute between both countries for its water share. The study area was divided into four zones, Sulemanki Zone, Islam Zone, Mailsi Syphone Zone and Panjnad Zone. Liquid Liquid Extraction (LLE) technique was used for the collected water samples followed by high performance liquid chromatography (HPLC) UV-Visible detector The current finding revealed that aldrin was not detected during summer period in water samples of SZ-1 (Sulemanki Barrage), SZ-2 and SZ-3 (Maisli Siphon) of the study area. Lindane and DDE were found more in the samples of sediments from the study area at SZ-4 ranging from 2.238-8.226 ppb and 4.234-6.876 ppb, respectively. Heaptachlor (in sediments) was found to be0.032-234 ppbonly at SZ-4.Endosulfan concentrations in water (winter) at SZ-3 was 0.06 ppb and at SZ-4, it was 0.05 ppb; dieldrin in water (winter) at SZ-4 was 0.0314 ppb and heptachlor was detected at SZ-1 (0.0315 ppb) and SZ-2 (0.0310 ppb) in water during winter season, were reaching to the Maximum Concentrations Limits

(MCL), while all other residues investigated were found below the MCLin all the compartments of the study area set by various agencies like WHO/FAO- Codex Alimenterious. Present findings revealed that although the organochlorine pesticides are banned for agricultural use in many countries, including Pakistan, their presence in various samples might be due to illegal use of these pesticides in the study area and its neighboring regions. The overall study area comprises of mainly urban, suburban and agricultural land being the largest cotton growing area of the country. There is a need to take serious steps to minimize water pollution caused by pesticides to achieve a healthy lifestyle.

#### VARIETAL PREFERENCE OF SUCKING INSECT PESTS ON MUSTARD VARIETIES

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The trial was conducted at Mustard Oil Field, Agriculture Research Institute (ARI), Tandojam in RCBD design. For this, 05 cultivars (UCD-1202, NMT-8, Canola, Nawab Shah and P-25) were screened out against the target pests. The pest monitoring was done at weekly interval. Highest infestation of whitefly (0.98±0.29 nymphs per plant) was noted for Nawab Shah followed by P-25 (0.92±0.26 nymphs per plant), NMT-8 (0.89±0.25 nymphs per plant) and Canola (0.82±0.22 nymphs per plant), while lowest infestation (0.77±0.21 nymphs per plant) was observed for UCD-1202. Highest infestation of thrip (3.20±0.78 nymphs per plant) was recorded for Nawab Shah followed by P-25 (3.08±0.75 nymphs per plant), NMT-8 (2.97±0.70 nymphs per plant) and Canola (2.93±0.6 nymphs per plant), while lowest infestation (2.50±0.57 nymphs per plant) was observed for UCD-1202. Overall mean highest population of jassid (0.74±0.20nymphs per plant) was recorded for Nawab Shah followed by P-25 (0.60±0.30 nymphs per plant), Canola (0.59±0.15 nymphs per plant) and NMT-8 (0.56±0.16nymphs per plant), while lowest infestation (0.19±0.05 nymphs per plant) was observed for UCD-1202. Highest infestation aphid (21.80±10.28 nymphs per plant) was recorded for Nawab Shah followed by P-25 (20.78±10.10 nymphs per plant), NMT-8 (20.48±9.64 nymphs per plant) and Canola (18.85±9.07nymphs per plant), while lowest infestation (18.27±9.04 nymphs per plant) was observed for UCD-1202. Maximum crop yield (1850.53 kg plot<sup>-1</sup>) was recorded for UCD-1202 followed by NMT-8 (1560.2 kg plot<sup>-1</sup>), P-25 (1540.5 kg plot<sup>-1</sup>) and Canola (1520.3 kg plot<sup>-1</sup>) and the minimum crop yield (1498.8 kg ha<sup>-1</sup>) was noted for Nawab Shah mustard variety. It is concluded that maximum infestation of whitefly, thrip, jassid and aphid was observed for variety 'Nawab Shah' and minimum was observed for variety 'UCD-1202'.

#### PERFORMANCE OF VARIOUS COLORED STICKY TRAPS AGAINST INSECTS OF MAIZE.

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The field study on relative performance of various colored sticky traps against insects of maize (*Zea mays* L.) was conducted during 2019. Five different colored sticky traps i.e., yellow, blue, green, white and transparent were installed, where each trap was replicated four times. The field was divided into four blocks, each having a size of 30,000 square feet. In each block, five colored sticky traps (white, yellow, green, blue and transparent) were randomly installed at one feet above the crop height. Weekly observations were taken to count the number of insects attracted to various colored from germination till harvesting of crop. Identification of insects was done on available literature. Data of abiotic factors were also obtained to determine their influence on attractiveness of colored sticky traps for insects. During the study, six insects i.e., aphids, leafhoppers, thrips, shootfly, mirid bugs and zigzag beetles

were found on various colored traps. The peak population of aphids was attracted on yellow and blue sticky traps, whereas, leafhoppers, shootfly, mirid bugs, and zigzag beetles were attracted to yellow and green colored traps. However, thrips showed attraction towards yellow and white sticky traps. The overall weekly observation showed that peak populations of aphids, thrips, mirid bugs, zigzag beetles were recorded on 31st March, 2019 on various colored traps. The maximum population of leafhoppers and shoot fly was recorded on 6th April and 5th May, respectively. The populations of aphids, leafhoppers, thrips, and zigzag beetles showed positive and significant weakly a-biotic factors (temperature, relative humidity, wind velocity) significantly affected on the performance of colored sticky traps to attract various insects. Based on the findings, yellow and blue colored traps should be used for the early detection and monitoring of maize insects.

# PEST MANAGEMENT OF CHILI CROP (CAPSICUM ANNUM L.) BY BOTANICAL EXTRACTS

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The aim of this research is to manage the efficacy of different types of bio-extracts on growth of chili crop (capsicum annum L.) and note their effectiveness against pest infestation as well as on the crops' health. The study was done from July to October 2019 in eastern region of Karachi. Aqueous extracts of mint, lemon, neem and garlic were applied in field before sowing and after germination of chili seed. The residues of these extractions were used as fertilizers in field. Results were indicated that garlic and mint extracts are more effective against majority of biting and chewing type insects pests and act as best repellent. Neem aqueous extract act as a deterrent against majority of soil insect pests as well as major chili crop pests whereas lemon extract showed least efficacy. Due to soil treatment with bio-extracts, fungal and bacterial diseases were not attacked on the crop. The overall results showed that bio-extracts enhanced the healthy growth of chili crop as well as they act as best bio-pesticides with anti-microbial and anti-fungal properties but seed borne diseases were observed in the study area. The use of botanical extracts in integrated pest management (IPM) is very important to decrease the hazardous effects of chemicals in controlling pest infestation as well as bio-extracts are eco-friendly in nature as to protect non-targeted organisms (Raoul, et.al., 2019: Mazhawidza, et.al., 2018). Plant extracts can provide economically feasible methods for encountering pest problems against hazardous chemicals in future.

# TOXICOLOGICAL AND BIOCHEMICAL EVALUATION OF CEDRUS DEODARA OIL IN COMPARISON TO CARBOSULFAN AGAINST ADULT MEALWORMS TENEBRIO MOLITOR (COLEOPTERA: TENEBRIONIDAE)

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The toxicity of deodar oil, a phytoinsecticide from the deodar tree (*Cedrus deodara*), was evaluated in respect to its inhibitory effect on cholinesterase and increases in glutamate pyruvate transaminase and glutamate oxaloacetate transaminase in adult mealworms (*Tenebrio molitor*) in comparison to the synthetic insecticide carbosulfan. Both insecticides were applied through feeding and lethalities (LC<sub>50</sub>) were calculated. Deodar oil was found to have about a twentieth in efficacy compared to carbosulfan. Plant extract from deodar oil can be used for control of mealworms and other insect pests.

# BIOLOGICAL AND ECOLOGICAL EFFECTS OF CHEWING LICE (PHTHIRAPTERA) ON MALLARD ANAS PLATYRHYNCHOS (ANSERIFORMES: ANATIDAE) IN SINDH, PAKISTAN

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Mallard Anas Platyrhynchos L. is a common duck and game bird is very important source of food energy, sports recreation, ecotourism and ecological bio-factor. It harbors a variety of parasites. It has been examined for its chewing lice species in the present work. During ectoparasitic studies total 17 mallard ducks were collected from different water bodies of Sindh, Pakistan. Birds were brought to Advanced Parasitology Research Laboratory (APRL) Department of Zoology, University of Sindh, Jamshoro. Birds were examin0065d for their chewing lice through fumigation and visual examination methods, the recovered specimen were dehydrated in graded series of ethanol cleared in clove oil and finally permanently in Canada balsam. Four chewing lice species were recovered on mallard duck including, Anatoecous icterodes (Nitzsch, 1818), Anaticola crassicornis (Scopoli, 1763), Trinoton querquedulae (Linnaeus, 1758) and Holomenopon leucoxanthum (Burmeister, 1838. The complete life cycle of these four species were observed on mallard, their quantitative parameters like prevalence, population abundance, physical effects on feather and seasonal variation of all species of chewing lice were studied in the present project.

# OLFACTORY RESPONSE OF CHRYSOPERLA CARNEA AGAINST DIAPHORINA CITRI AND THEIR HOST PLANT

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Green lacewing, Chrysoperla carnea (Stephens) (Neuroptera: Chrysopidae) is a generalist predator. It predates on various stages of insect pest i.e. eggs, larvae, nymphs and adult. Citrus is an important fruit crop and damaged by several insect pest i.e. citrus psylla, scales, whiteflies, mealybug etc. Citrus psylla, Diaphorina citri (Kuwayama) (Hemiptera: Aphalaridae) is a severe pest of citrus and a vector of citrus greening. Feeding of D. citri induce the emission of volatiles from plants. Response of C. carnea towards plants infested by its prey is very critical. To now, no work has been performed to check the tri-trophic interaction i.e. citrus-citrus psylla-green lacewing. There is way to find the role of volatiles in the attraction of C. carnea towards plant and D. citri. Four arm olfactometer was used to check the role of volatiles released by plants in response of D. citri feeding. Different plant parts (new leaves, old leaves, fruit peel, and fruit) were used to check the response of C. carnea on plant and D. citri. These plant parts (undamaged and damaged by D. citri) were used in different combinations in olfactometer. In all bioassays, odor source was compared with combination, individual plant part and with control. When un-infested plant parts were given, the highest response was observed towards new leaves, followed by old leaves. While, when damaged plant parts were given similar but comparatively higher response was observed towards new leaves and old leaves. Whereas, minimum attraction was observed on fruit. These results suggested that, during host finding C. carnea use volatiles released by plants in response of D. citri feeding. Future work should comprise on the identification of plant volatiles induced by *D. citri* feeding and pheromones of *D. citri*.

# PREDATION STUDY OF CHRYSOPERLA CARNEA S. (NEUROPTERA: CHRYSOPIDAE) ON SPODOPTERA LITURA F. (LEPIDOPTERA: NOCTUIDAE) AT DIFFERENT TEMPERATURES

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Green lacewing, Chrysoperla carnea S. (Neuroptera: Chrysopidae) is a good predator of Spodoptera litura F. (Lepidoptera: Noctuidae) eggs and 1<sup>st</sup> instar larvae. C. carnea feeds on the eggs of S. litura with hairs, without hairs and 1<sup>st</sup> instar larvae. Five different temperatures (15°C, 20°C, 25°C, 30°C and 35°C) were used to check the predation of C. carnea. Result showed that predation was increased with increased of temperature and also from lower instar to higher instar. The maximum predation was found at 35°C, 3<sup>rd</sup> instar with hairs, without hairs and larvae (14.6, 15.6 and 15.8) and minimum predation was found 15°C, 1<sup>st</sup> instar with hairs, without hairs and larvae (2, 2 and 1.4). It is concluded that 35°C is an ideal temperature to control the pest and could be used in IPM strategies.

# SURVEILLANCE AND DOCUMENTATION OF FRUIT FLIES PARASITOIDS FROM INFESTED FRUITS OF DIFFERENT CLIMATIC ZONES OF THE SINDH

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Fruit flies belong to family (Tephritidae-Diptera) are the most injurious fruit pests of the fruits and vegetables. Biological control is an environmentally friendly control method that has been used against pest fruit flies. Parasitoids have been one of the most widely used groups of natural enemies. In present studies we have surveyed the biological agents from two different zones Sindh. The field locations were Hyderabad and Larkana districts from where infested fruits were collected from guava and mango orchards. The results revealed that significantly (P<0.05) higher number of larval cum pupal parasitoid *Trybliographa daci* were (62.2±3.03,50.20±4.45) recorded in guava from both districts. Furthermore, Maximum number of *Bactrocera zonata* infestation were recorded (395.6±4.50, 288.00±11.57) from guava orchards of both districts. This study established that Larval/pupal parasitoid *Trybliographa daci* proved most promising bio-agent in limiting the population of fruit flies in guava and guava observed most susceptible host for fruit flies in terms of infestation.

# EFFECT OF DIFFERENT ROW SPACING ON THE POPULATION OF APHID (APHIS CRACCIVORA KOCH.) IN CHICKPEA

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In order to investigate the effect of row spacing on the population of aphid (Aphis craccivora Koch.) in chickpea field of Pulse sub- station Agriculture Research Institute Tandojam, during Rabi season 2018-19. The

analysis of data showed that effects of row spacing were significant (P<0.05) on population of aphid. The results indicated that aphid appeared during the last week of November up to 1st week of March when the crop was harvested. In initial stage of chickpea crop during (15cm space) the population of the pest remained low with (1.57) during 1st observation (last week of November) afterwards it started building up the population reached to its peaks i.e., (20.73) during 2nd week of February. Then, the population was gradually declined with (15.83) during 1st week of March (last week of observation). The seasonal population mean was 12.28±1.89. Similarly, population build up was also observed on chickpea during (30 cm row space). However, during last week of November (1st observation) the population noted was (1.47), afterwards the population of the pest was increased to its peak i.e., (16.37) during 3rd week of February. Then after population of aphid decreased with (15.83) during 1st week of March (final observation). The seasonal population mean was 8.94±1.37. After that population of pest was recorded (45cm row space) with initial observation (Last week of November with (2.50) per plant population. The population reached to its maximum (12.80) during 2nd week of February. Thereafter, its population declined and reached to (7.87) up to harvest of crop in 1st week of March. The seasonal population mean was 8.03±1.24. While correlation of abiotic factors reveled that temperature and relative humidity had negative relation with aphid population.

### STUDIES ON THE PREVALENCE OF *HELICOVERPA ARMIGERA* ON TOMATO CROP IN DISTRICT TANDO ALLAHYAR SINDH

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Present study was conducted in district Tando Allahyar from August 2018-April 2109. The study was conducted at Dero agriculture form where two type of forming selected (Tunnels forming and without tunnels traditional forming), and village Umar Kakepota each comprising on one hector area. Infestation of *H. armigera* was observed in both types of forms, without tunnels traditional form and Tunnels form. Infestation rate was observed high in traditional forming than Tunnels forming. During the current study the infestation in the both types of farming was seen high from September to October 2018, while as the higher rate of infestation was calculated in September 23.5% in traditional form while 7.5% infestation recorded in tunnels form during the month of September. While at Village Umar Kakepota the peak infestation was recorded in the month of October 23% (without tunnels). Present study revealed that tunnels forming of tomatoes is latest technology based forming so infestation tomatoes recorded less than traditional farming. *Helicoverpa armigera* was observed the major tomato fruit and bud borer in studied locality.

# PROTECTIVE EFFECTS OF PLANT EXTRACTS AGAINST DELTAMETHRIN INDUCED TOXICITY IN RATS

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A study was carried out to evaluate changes occur in albino rats due to deltamethrin induced toxicity and to know about protective effects of two antioxidants (*Moringa oleifera* and *Bougainvillea spectabilis*) against pesticidal effects. Experiment were carried out for 30 days on twelve healthy male albino rats in Animal House of Food Science and Technology department of Agriculture University Faisalabad. Rats were acclimatized for one week and kept under favorable condition. All of these rats divided into 4 groups. Go was taken as control group that fed on simple diet and water receiving no chemicals and plant extract. G1 received daily dose of deltamethrin (1.55 ml) along with *bougainvillea spectabilis* extract (150 mg) per body weight orally. G2 received deltamethrin (1.56 ml) along with

Moringa oleifera extract (150 ml) and G3 received deltamethrin only about 1.57 ml. Each rat acted as individual replica. Daily dosage of mixture was offered to all these groups. Autopsy of these rats was done after a considerable period to analyze certain changes in all hematological parameters. RBC, MCV, HCT, HGB, MCH, MCHC and LYM significantly (P<0.01) decreased when deltamethrin was given but increased when antioxidants were given along with the pesticide. However, WBCs increased significantly with deltamethrin administration and decreased when rats were treated with two antioxidants. The study proved that both plant extracts can ameliorate toxic effects of toxicants in mammals.

# LETHAL AND SUB-LETHAL DOSE RESPONSE OF INSECTICIDES AGAINST PINK BOLLWORM (P. GOSSYPIELLA) FOR ITS SUSTAINABLE MANAGEMENT

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Cotton is one of the most significant crops responsible for the production of natural fiber, also called "White gold" or "King of fibers". Pakistan is the fourth cotton producing country in the world. Cotton production is badly affected due to attack of various sucking and chewing insect species. Among chewing pests, Pink bollworm, (Pectinophora gossypiella) is the major damaging pest of this crop, and is destroying the crop at larval stage because of chewing mouth parts. Increasing resistance in pink bollworm due to improper and non-compatible use of insecticides is main problem facing by farmers. The main objective of current study was to evaluate the efficacy and suitable combination of different insecticides to manage the pink bollworm (P. gossypiella) population using different lethal, sub-lethal concentration alone and in combination. Present experiments were conducted to observe the effectiveness of spinosad, cypermethrin, coragen. Three levels of each insecticide were used. All the treatment alone and in combinations was incorporated into the artificial diet for feeding. Results indicated that combination of lethal dose of coragen and sublethal dose of spinosad observed maximum larval mortality (96%) showing synergistic interaction. While lethal dose of deltamethrin with lethal and sublethal doses of coragen showed no change or antagonistic interaction with mortality 40.34% and 32 % respectively. It is concluded that coragen having chlorentraniliprole active ingredient in combination with Spinosad is best for the management of P. gossypiella.

# SEASONAL PREVALENCE OF *P. SOLENOPSIS* ON OKRA CROP IN DIFFERENT REGIONS OF SINDH PROVINCE

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The surveys were conducted at different locations of Sindh viz; ., Digree, Hala, Jhudo, Kot Ghulam Muhammad, Kotri, Mirpur Khas, Mirpur Sakro, Mitiari, Noukot, Sakrand, Sanghar, Sarhari, Sheikh Bhirkio, Sultanabad, Tando Adam, Tando Muhammad Khan, Tando Qaisar, Tando Allahyar, Tando Jam, Usman Shah, Nasarpur, Chamber, Bukera Shareef, Qubba Stop, Kamaro Shareef, Jhando Mari, Khuwaja Village, Bhatta stop, Detha, Rahoki, Kissana Mori, Hosri, Mokh Shareef, Pir Kathi, Nawab Shah and Began Jarwar during the months of June 2014 to November 2015. The present study reveals that the highest mealy bug population was found at Mirpur Khas 188.95 + 4.9, 183.7 + 4.16 and Sheikh Bherkio 18.3+4.16 during July 2014. On the other hand, there was no

mealy bug population has been recorded during the month of January and February 2015. Only single species of mealy bug, *Phenacoccus solenopsis* was found at all okra growing regions. After the surveying five different districts sites such as Tandojam, TandoAllahyar, Sulatanabad, Mitiyari and Sakrand were selected for studying the P. *solenopsis* population dispersion. The maximum population of P. *solenopsis* were notified at  $2^{nd}$  half of August and  $1^{st}$  of half of September 2015 at all studied districts. Generally, it appears that okra mealy bug population was positively correlated with the temperature and relative humidity.

# PARASITISM, EMERGENCE AND POST EMERGENCE SEX RATIO OF *DIRHINUS GIFFARDII* (SILVESTRI) (HYMENOPTERA: CHALCIDIDAE) AGAINST THE PUPAE OF *BACTROCERA CUCURBITAE* (COQUILLETT) (DIPTERA: TEPHRITIDAE) AT DIFFERENT EXPOSURE PERIODS

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Pupal parasitoid, *Dirhinus giffardii* (Silvestri) is an significant biological control agent that has been used effectively in controlling the tephritid flies, including *Bactrocera cucurbitae*. Fruit flies being severe pest of fruits and vegetables are responsible for enormous economic losses in the world. Laboratory studies were conducted to investigate the parasitism, emergence and post emergence sex ratio of *D. giffardii* through different exposure times and parasitoid density against the pupae of melon fruit fly, *B. cucurbitae*. The studies manifested that the exposure time and parasitoid density had a significant effect on the parasitism and emergence ratio of parasitoids. Results revealed that mean rate of parasitism and mean rate of emergence of *D. giffardii* was recorded highest in case of five pair of parasitoids and exposure period of six days. Further, it was observed that, exposure time and parasitoid density had no significant effect on the post emergence sex ratio of male and female parasitoids. The mean per female parasitism was increasing with the increase in number of pairs of *D. giffardii* and reached to its peak after exposure time of six days. These findings suggest that *D. giffardii* has a great parasitizing potential against the pupae of *B. cucurbitae* and can effectively suppress its populations.

# IMPROVED DELIVERY SYSTEM OF BEAUVERIA BASSIANA THROUGH DEVELOPMENT OF FOAM FORMULATIONS FOR MANAGING BACTROCERA ZONATA

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Bactrocera zonata Saunders (Tephritidae: Diptera) is one of the biggest constraints for mango production in Pakistan. It deteriorates quality and quantity due to premature dropping. Yellow warning issued to Pakistan in 2013 due to Fruit fly in export shipments. The presence of maggots in the mangoes make it rejected and the greatest threat to make it unfit for the consumption of human. Present study was focus on the colour preference of B. zonata, evaluating pathogenicity of entomopathogenic fungus, Beauverai bassiana. Further the delivery of B. bassiana was improved by applying it in foam-mixed formulation. The study was conducted at Muhammad Nawaz Shareef University of Agriculture, Multan and mango orchid of Mango Research Institute, Multan in three replicates. Pathogenicity of B. bassiana was tested on larval (L2), pupal and adult stage of B. zonata. Growth and development (larval duration, pupal duration, adult longevity), mortality, pupation, adult emergence, eclosion, mycosis and sporulation was recorded. Our findings showed that B. zonata prefer full yellow colored traps (73.09±2.84 flies/trap/week), whereas a little preference was noted for traps with green lid and yellow bodied traps (45.92±2.00 flies/trap/week). The present investigation uses four different colour combination of fruit fly traps viz., full green, full yellow, yellow lid/green body, green lid/yellow body. Role of treatment contrast with the population and its mortality different four treatments have a different result like fungus+Foam (B. bassiana) shows higher mortality

63.05±2.8688, as compare to the last fourth treatment which is untreated or controlled. Therefore, timely installation of these traps is recommended as a part of integrated pest management for monitoring, detection and control of fruit flies.

# FUNCTIONAL RESPONSE OF THE PUPAL PARASITOID, DIRHINUS GIFFARDII TOWARDS TWO FRUIT FLY SPECIES, BACTROCERA ZONATA AND B. CUCURBITAE

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Functional response is usually used to evaluate the potential of a parasitoid or predator in regulating the population of a pest and hence, is an important tool in the selection of appropriate biological control agents for a particular pest. This study aimed to investigate the functional response of the parasitoid, *Dirhinus giffardii* Silvestri (Hymenoptera: Chalcididae) towards two *Bactrocera* species (Diptera: Tephritidae), *B. zonata* (Saunders) and *B. cucurbitae* (Coquillett); each offered at seven different densities (10, 20, 30, 40, 50, 70 or 100 pupae) for a period of 24 h in cylindrical plastic cages in the laboratory. The results showed a type II functional response of *D. giffardii* towards both the fruit fly species, *B. zonata* and *B. cucurbitae*. The parasitoid exhibited a higher attack rate (a) (0.8235 vs. 0.6798), a shorter handling time ( $T_h$ ) (0.05160 vs. 0.7344) and a higher maximum parasitism rate ( $T_h$ ) (46.52 vs. 32.69) on the pupae of *B. zonata* than on the pupae of *B. cucurbitae*. Consequently, the parasitoid was more effective on *B. zonata* than on *B. cucurbitae*. Offspring sex ratio of the parasitoids was female biased regardless of the host fruit fly species. The results suggest that *D. giffardii* could be a more efficient biological control agent for *B. zonata* than for *B. cucurbitae*.

#### IDENTIFICATION OF THRIPS IN MANGO NURSERY, MULTAN

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Mango (Mangifera indica: Anarcardiaceae) is the major fruit crop of Multan, Pakistan. It is very demandable due to its sweetness, flavor, texture and nutritional qualities. That's why it is called king of fruits. About 1% of total production used in making of juices, jams, jellies etc. True nursery plants are responsible for healthy mango trees in future. They are very delicate in nature and effected by various environmental factors throughout the year. Among different factors insect pests mainly thrips gaining importance by the passing of time. It causes serious loss to mango nursery plants by sucking cell sap in gregarious forms. Young mango leaves are mostly affected and they cannot able to grow well. It also act as vector of tospovirus. Continues damage by thrips on mango nursery plants insisted to know it actual morphology for effective management. For thrips its genus was studied by using key. The results indicate that it belongs to suborder terebrantia because its last abdominal segments are conical in shape. Ctendium are present on tergal segments and antenna is 8- segment. 1 pair of post ocular setae present on the head. These indication exhibits it belongs to genus *Scirtothrips sp.* 

# EFFECT OF ROW SPACING ON THE POPULATION OF MUSTARD APHID (*LIPAPHIS ERYSIMI* KALT.)

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In order to investigate the effect of row spacing on the population of aphid (Lipaphis erysimi Kalt.) in mustard field of Entomology Section, Agriculture Research Institute Tandojam, during Rabi season 2018-19. The analysis of data showed that effects of row spacing were significant (P<0.05) on population of aphid. The results indicated that aphid appeared during the first week of December up to last week of February when the crop was harvested. In initial stage of mustard crop during (30cm space) the population of the pest remained low with (5.47) during 1st observation (first week of December) afterwards it started building up the population reached to its peaks i.e., (20.90) during last week of January. Then, the population was gradually declined with (8.03) during last week of February (last week of observation). The seasonal population mean was 13.89±2.31. Similarly, population build up was also observed on mustard during (45 cm row space). However, during first week of December (1st observation) the population noted was (6.46). Afterwards the population of the pest was increased to its peak i.e., (16.53) during 3<sup>rd</sup> week of January. Then after population of aphid decreased with (8.00) during 3<sup>rd</sup> week of February (final observation). The seasonal population mean was 10.64±1.77. After that population of pest was recorded (60cm row space) with initial observation (1st week of December with (4.23) per plant population. The population reached to its maximum (12.93) during 2<sup>nd</sup> week of December. Thereafter, its population declined and reached to (10.13) up to harvest of crop in 3rd week of February. The seasonal population mean was 9.80±1.63. While correlation of abiotic factors reveled that temperature and relative humidity had negative relation with aphid population.

### MANAGEMENT OF GUAVA FRUIT FLY SPECIES AT DIFFERENT FARMS SURROUNDING LARKANA DISTRICT

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The studies on management of guava fruit fly species at different guava farms of Rafique Bhutto, Hamid Rattar, Hameed Jessar and Muqeem Jessar were conducted near Larkana city from October 2016 to March 2017. The results of weekly catches of fruit flies through lure traps (T1) and lure + monitor pesticide (T2) indicated the presence of two species of fruit flies, bactrocera (Dacus) zonata (zonatus) (saunders) and Bactrocera (Dacus) Dorsalis (handle) in the experimental area. The highest number of fruit fly, Bactrocera zonata ranged between (4507 to 555) / five traps / week 4507, 3649, 3337, 1447 and 1083 during 1st week of October, 3rd week of November, 2nd week of December, 4th week of January, and 1st week of February 2017, respectively indicting five generation throughout the period under study. The catches adult of fruit fly, Bactrocera dorsalis indicated that the adult population was minimum with a range of 1 to 8 / five traps in both in treatments. The seasonal mean / traps was 0.48 and 0.56 in T1 and T2, respectively. The adult catches in T2 (Lure + pesticide) indicated population range of (261 to 2232) / five traps throughout fruiting season. The adult population catches were oscillating almost after every month with peaks of 2211, 2232, 1948, 1808 and 1177 in 1st week of October, 4th week of November, 2nd week of December, 4th week of January and 4th week of February. Thereafter, the population disappeared during 1st week of March due to the harvest of guava fruits. The seasonal mean catches of adult fruit flies trap were highest 323.52 in T1 (lure traps) than T2 (lure + pesticide) with 238.41 / trap. The lowest seasonal mean catches / traps (23.48) in lure traps + pesticide indicated that monitor pesticide with lure trap was more effective in reducing the pest.

# INFLUENCE OF THREE APHID SPECIES FOR DETERMINING THE STAGE-SPECIFIC FUNCTIONAL RESPONSE OF *MENOCHILUS SEXMACULATUS* (COLEOPTERA: COCCINELLIDAE)

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Coccinellids are common predators of aphids including *Aphis nerii*, *Diuraphis noxia*, and *Lipaphis erysimi*. Zigzag ladybird beetle, *Menochilus sexmaculatus*, is very effective against various aphid species. In the present study different stages of *M. sexmaculatus* were evaluated for their functional response towards *A. nerii*, *D. noxia*, and *L. erysimi*. Functional response at different densities of aphids was calculated over a time of 24h at 25±5°C and 65±5% RH. Roger's Random Predator equation was used to calculate parameters of functional response. All the predatory stages showed functional response of Type II. Attack rate (*a*) was highest in fourth instar for all the aphid species (4.5171, 2.5894 and 2.8853, respectively), whereas handling time (T<sub>h</sub>) of fourth instar was recorded minimum for *A. nerii* (0.0175h) and *L. erysimi* (0.0159h). In case of *D. noxia*, minimum handling time was observed for first instar (0.0175h). Fourth instar of *M. sexmaculatus* consumed more prey followed by third instar. So, these results indicates that 4<sup>th</sup> instar of *M. sexmaculatus* was more predacious followed by third instar larvae, adult female and male. These predatory stages are more suppressive against these three aphid species.

# RELATIVE PERFORMANCE OF DIFFERENT COLORED STICKY TRAPS AGAINST *THRIPS TABACI* L. ON ONION

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Thrips tabaci (Lind.) is one of the most important insect pests which damage all stages of onions. Therefore, present study was conducted at experimental field, Entomology Section, ARI, Tandojam during 2018 to determine the influence of various colored sticky traps in the attraction of *T. tabaci*. The Phulkara variety of onions was used in the study as its nursery was obtained from the Onion Research Station, Husri. Five colored sticky traps i.e., blue, green, yellow, white and transparent were used in the study. Observations were taken from both sticky traps and onion plants of the respective colored sticky trap treatments on weekly basis. Experiment was arranged in a RCBD design with four replications. Results showed that among sticky traps, blue color sticky trap were found to be significantly more effective in attracting T. tabaci (39.49 $\pm$ 1.60 thrips/sticky trap), followed by yellow sticky traps (32.66±1.27 thrips/sticky trap). Moreover, green, white and transparent were found less attractive for T. tabaci with attraction level recorded at  $15.37\pm0.75$ ,  $8.27\pm0.47$  and  $3.20\pm0.19$  thrips/sticky trap, respectively. It has been also recorded that on onions plants, significantly higher number of T. tabaci (24.621±1.080 thrips per plant) was observed transparent colored sticky trap treatment, followed by white (22.92±0.930 thrips per plant) and green (21.951±0.94 thrips per plant) sticky trap treatments. The lowest number of T. tabaci population on onion plants were recorded on blue sticky trap (19.369±0.99 thrips per plant) treatment. Considering the maximum attraction of thrips to blue sticky traps, thus lowering its population on onion plants, the highest onion yield was recorded in blue trap treatment (80.33+1.73 mds/acre), whereas, the lowest yield was recorded in transparent sticky trap treatment (64.67+2.00 mds/acre). Therefore, it is concluded that blue sticky traps should be considered for the better monitoring and as a component of integrated management of T. tabaci.

# NATURAL ENEMIES OF RHYNCOPHORUS FERRUGINEUS (OLIVIER) IN DATE PALM AT KHAIRPUR AND POSSIBILITIES OF BIOLOGICAL CONTROL IPM OF THE PEST

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First time in Pakistan, survey was carried out for natural enemies associated with red palm weevil Rhyncophorus ferrugineus (Olivier). The survey was confined to Khair Pur district, the most date palm growing area of Sindh province of Pakistan. Out of survey conducted in Khair Pur 13 species of natural enemies were recorded that attacked red palm weevil (RPW) larvae, pupae and adults. Three of these belonged Arachnida (mites Uroobovella marginata (Koch.), Aegyptus alhassa Al-dhafar & Al-Qahtni and Centrouropoda almerodai Hiramatsu and Hirschmann); one belonged Diptera {Megaselia scalris (Loew)}, one Formicidae (Camponotus pennsylvanicus De Geer); one Dermoptera (ear wig) and one Carabid. In addition, predatory birds Dendrocitta vagabunda (Latham), Corvus splendens Vieillot, Bubulcus ibis (L.), Acridotheres tristis (L.) and Turdoides striata (Dumont) and a fungus disease Beauveria bassiana (Bals.) were recorded. Identifications of natural enemies were obtained from Natural History Museum, London, UK; CABI, Rawalpindi and Subject Specialist Taxonomist, Agriculture University, Faisalabad. Overall mortality due to fungus disease, insects and mites was very low in most of the cases, however, in some cases it was high up to 80%. RPW adults were caught in pheromone traps in more or less numbers throughout the year. Besides survey for natural enemies' possibilities were explored for their use in controlling RPW in date palm. Experiments were conducted on augmentation and conservation of some promising predatory mites, Uroobovella marginata. and Centrouropoda almerodai. Cultures of these mites' species were established in the laboratory and released in an orchard of the infested date palm trees. On each tree, approximately 10,000 *U. marginata* individuals were released in June 2019 and data was recorded on monthly basis for consecutive three months to check if this had any role in destroying RPW larvae and pupae. High mortality of RPW larvae and pupae was recorded in range of 79.1-80.8%. Improved cultural practice of cutting dying infested trees into pieces and keeping them exposed in field for 72 hours provided the chance of massive predation of RPW by birds and other generalists. Options of increased mortality of RPW by augmentation of predatory mites; year-round catch of RPW adults in pheromone traps and improved cultural practice of cutting young dying trees into pieces for massive predation of RPW larvae by birds provided strong foundation to further work on integrated approach of combining the different techniques for controlling RPW.

# COMPARATIVE DAMAGE ASSESSMENT OF PULSE BEETLE CALLASOBRUCHUS MACULATUS ON DIFFERENT PULSES IN CHOICE CONDITIONS

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Pulse beetle *Callasobruchus maculatus* is a worldwide major pest of pulses. Mainly larvae cause damage to pulses by feeding inside the grains. However, a significant impact of composition of pulses has been recorded in the feeding, oviposition and development of *C. maculatus*. Therefore, this study was undertaken to determine influence of various major pulses on population development and damage of *C. maculataus*. Five pulses i.e., grams, masoor, dall channa, chaunra and mung was used in the study. A standard weight of 100 grams of each pulse was put in separate boxes that were attached together. Twenty pairs of *C. maculatus* were released at the center of the boxes to give beetles equal chance to select their preferred pulse. Afterwards, weekly observations were recorded for two months to record for the population growth of adults along with number of eggs laid. Weight loss was recorded at the end of experiment. Results indicated that overall, maximum population of *C. maculatus* eggs was recorded in chaunra (110±21.70 eggs), whereas, minimum population was observed in masoor (11.50±3.10 eggs), followed by grams (19.50±5.70 eggs). Similarly, the highest male and female population was also recorded in chaunra, followed by mung. Considering the population of beetles, overall, the highest and lowest weight loss was recorded in chaunra, mung and masoor, respectively. Therefore, considering the variable preference and losses of *C. maculatus* to various

pulses, it is suggested that proper arrangement should be done to store pulses adequately in separate places to restrict the movement and losses *C. maculatus*.

# MANAGEMENT OF GUAVA FRUIT FLY SPECIES AT DIFFERENT FARMS SURROUNDING LARKANA DISTRICT

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The studies on management of guava fruit fly species at different guava farms of Rafique Bhutto, Hamid Rattar, Hameed Jessar and Muqeem Jessar were conducted near Larkana city from October 2016 to March 2017. The results of weekly catches of fruit flies through lure traps (T1) and lure + monitor pesticide (T2) indicated the presence of two species of fruit flies, bactrocera (Dacus) zonata (zonatus) (saunders) and Bactrocera (Dacus) Dorsalis (handle) in the experimental area. The highest number of fruit fly, Bactrocera zonata ranged between (4507 to 555) / five traps / week 4507, 3649, 3337, 1447 and 1083 during 1st week of October, 3rd week of November, 2nd week of December, 4th week of January, and 1st week of February 2017, respectively indicting five generation throughout the period under study. The catches adult of fruit fly, Bactrocera dorsalis indicated that the adult population was minimum with a range of 1 to 8 / five traps in both in treatments. The seasonal mean / traps was 0.48 and 0.56 in T1 and T2, respectively. The adult catches in T2 (Lure + pesticide) indicated population range of (261 to 2232) / five traps throughout fruiting season. The adult population catches were oscillating almost after every month with peaks of 2211, 2232, 1948, 1808 and 1177 in 1st week of October, 4th week of November, 2nd week of December, 4th week of January and 4th week of February. Thereafter, the population disappeared during 1st week of March due to the harvest of guava fruits. The seasonal mean catches of adult fruit flies trap were highest 323.52 in T1 (lure traps) than T2 (lure + pesticide) with 238.41 / trap. The lowest seasonal mean catches / traps (23.48) in lure traps + pesticide indicated that monitor pesticide with lure trap was more effective in reducing the pest.

# RESISTANCE ASSESSMENT IN RHYZOPERTHA DOMINICA (COLEOPTERA; BOSTRICHIDAE) TO PHOSPHINE AND DELTAMETHRIN UNDER LABORATORY CONDITIONS

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The lesser grain borer *Rhyzopertha dominica* (F.) is the most destructive pest of the stored grains and responsible for causing the tremendous loss to stored grains depend upon the storage structures and conditions. It deteriorates the quality and quantity of the stored grains. Most of the insects are build-up their populations during hot and humid environment. This insect pest is controlled by the application of Pyrethroids insecticides like deltamethrin and fumigants like phosphine gas. The current study was carried out to monitor the level of resistance in lesser grain borer against phosphine and deltamethrin. There were six concentrations of phosphine (0.006, 0.007, 0.008, 0.009, 0.01 and 0.011 ppm) and deltamethrin (0.01, 0.02, 0.03, 0.04, 0.05 and 0.06 ppm) were used against this test insect. The mortality data of this insect against phosphine was recorded after 7, 14 and 21 days' time interval. The data regarding mortality (%) of this insect against deltamethrin was recorded after 24, 48 and 72 hours. Phosphine gas was generated from aluminum phosphide tablets. Fumigation chamber (10.8 L) were used for the phosphine application. Completely randomized design (CRD) was used for this experiment under laboratory conditions. All the treatments were replicated three times. The results of the current study showed that the maximum mortality of resistant and susceptible strain of *Rhyzopertha dominica* against phosphine at 0.011 ppm was 56.35% and 31.15% respectively.

All the data showed that  $Rhyzopertha\ dominica$  was 3.65 times fold resistant against deltamethrin after 72 hour time interval than over susceptible strains. The LC<sub>50</sub> value showed that  $Rhyzopertha\ dominica$  was 0.882 times fold resistant against phosphine after 7 day's interval than over susceptible strains.

# EFFECT OF TEMPERATURE ON POPULATION DYNAMICS OF MANGO SCALES IN TEHSIL JATOI DISTRICT MUZAFFARGARH

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Mango is an important fruit of tropical and subtropical regions and has got the position of king of fruits due to its nutritious value and sweet taste. Mango production in Pakistan is declining because of various insect pest attacks. Among many mango pests, mango scales are gaining importance as these are present on the upper and lower side of the leaves and fruits and suck the cell sap of the leaves and other tender plant parts and reduce the vigor of the plants. Leaves infested with scale insects turn pale green or yellow and ultimately subjected to death. Due to its heavy infestation leaves and branches may also be killed and growth and fruit bearing capacity of tree is adversely affected. Both the quantity and quality of mango fruit is affected by scale insects by causing blemishes that affects the commercial value of fruits. A study was conducted on population dynamics of scale insects to assess the presence in mango orchard from April to December, 2016 in Tehsil Jatoi, District Muzaffargarh. Data was recorded on fortnightly basis from Chaunsa mango cultivar. Results revealed that initial mean population 0.56 was observed during the early weeks of April then gradually increased and reached up to the maximum mean population 1.60 in first fortnight of August. Then its mean population decreased to its lowest level 0.10 during first fortnight of November. It was concluded that mango scale population showed positive correlation with mean average temperature.

### EVALUATION OF INSECTICIDES FOR MANAGEMENT OF MANGO FRUIT BORER

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Mango fruit borer is a pre-dominating insect pest of mango. It has recently appeared in serious pest in mango growing areas of the Punjab, Pakistan. It is causing 10 to 52% damage of fruits from pinhead stage to full maturity. It is likely to spread in the new areas such as Bahawalpur, Rahim Yar Khan and lower Sindh. The insect has five larval instars and it passed the off-season as pre-pupae inside the cracks and crevices of the tree. Damaged fruits became unfit for human consumption. Present study was conducted to evaluate the efficacy of different chemical insecticides against Mango Fruit Borer. By the Application of Bifenthrin, Radiant, Emmanectin Benzotae, Luefenoron, Coragen and Voliam Flexi, Our results depicted that Bifenthrin was provided effective results for Mango Fruit Borer. It was observed that Mango Fruit Borer population was highest in Sindhri cultivar as compared to Black Chaunsa, Anwar Ratool, White Chaunsa, Fajri, and Langra. Our results concluded that Sindhri cultivar was most susceptible against Mango Fruit Borer.

# GLOBAL THREAT TO DATE PALM PLANTATIONS: RED PALM WEEVIL RHYNCHOPHORUS FERRUGINEUS OLIVER BIOLOGY AND MANAGEMENT TACTICS

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Date palm plantations across the globe are threatened by Red Palm Weevil, *Rhynchophorus ferrugineus* Oliver (Coleoptera: Curculionidae). Losses to date palm plantations in Pakistan sometimes surpass 10%-20%. Most of the traditional management strategies used by farmers have been found insignificant to combat this voracious pest. This review covers the history and spread of *R. ferrugineus* infestation across the globe, its economic impact and life history. In addition, the latest techniques for eradication of the pest and current management practices to protect date palm plantations against the weevil. The management options that have been considered to control *R. ferrugineus* include: early detection methods (acoustic sensors, visual and thermal imaging), use of pheromones, (monitoring and mass trapping), sterile insect technique (reproductive control), and insecticides, and biological control (natural enemies such as entomopathogenic fungi, bacteria etc.). Considering the complexity of the *R. ferrugineus* life cycle, integrated pest management offers an effective control strategy to address the pervasive pest.

#### EVALUATION OF DATURA METEL AGAINST BLACK RAT, RATTUS RATTUS

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Datura, Datura metel, L. is a toxic wild shrub which is found extensively in the tropical regions of the world. Commonly known as thorn apple devil's trumpet and metel. All parts of *Datura* plant are highly poisonous and may be fatal if ingested by humans or other animals, including livestock and pets. Datura metel may be toxic if ingested in a tiny quantity, symptomatically expressed as reddened skin, headaches, hallucinations, and possibly convulsions or even a coma. The major toxic compounds are terpenoids and alkaloids. This toxic plant may be utilized for rodent management by the farmers. In this regard a study was undertaken to evaluate the toxicity of flowers of Datura metel against black rat, Rattus rattus. The rats were trapped from a local grain market. The rats of approximate same size were sexed, weighed and caged individually in laboratory for 15 days. Ethanolic flower extract of Datura metel was tested in three doses (0.50%, 1.00% 2.00%, 5.00%) against the rat. Datura bait was formulated by mixing Datura extract (accordingly as per dose) in 1000g wheat flour and broken rice (equal quantity) and 2.00% brown sugar (as taste additive). For comparative evaluation (control) bait without Datura was prepared with a mixture of wheat flour, broken rice and brown sugar. Twenty rats (10 male & 10 Female) were weighed, caged individually and starved for four hours (before the start of each test). Twenty gram bait was given to each rat for each concentration for five days. Bait eaten (g), behavior and health of the rats were monitored continuously and mortalities were recorded. The bait proved it palatable; however, the bait acceptance was inversely proportional to the dose offered. All the doses proved very effective against the rats. The resulting intoxication caused reduction of feed consumptions and triggered tachycardia, difficulty in breathing, shakings, indigestion, anxiety, fatigue, muscular tremor, loss of muscle coordination and ruffled hair coat in the rats. Datura metel gave 100% mortality in 4.50± 1.12 days at 5.00%, 75% mortality in 6.65± 1.32 days at 2.00%, 40% mortality in 8.00 ± 2.20 days at 1.00% and 10% mortality in 8.00±1.25 days at 0.50% concentrations. In control group all the rats survived and exhibited normal behavior. This is a preliminary study however after some more studies the use of this easily available wild plant may be suggested for rodent control. These findings may be a step towards the development of safe, economical and eco-friendly rodenticides by utilizing bio-active plants as a substitute of conventional rodenticides.

# SCREENING OF RICE (ORYZA SATIVA L.) GERMPLASM AGAINST RICE LEAF FOLDER (CNAPHALOCROCIS MEDINALIS G) UNDER GREENHOUSE CONDITION

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Rice leaf folder (*Cnaphalocrocis medinalis* G.) is a key insect pest of rice crop (*Oryza sativa*) and distributed all over the rice growing areas of the world including Pakistan. The infestation of rice leaf folder often appears very suddenly resulting in huge yield losses. Host plant resistance is an important component of IPM. It is easy to use, viable, durable, effective and long term method as compared to any other control measure. It was imperative to exploit the sources of resistance to manage the pest and is environmental friendly control method. Present study was carried out in greenhouse of Plant Breeding and Genetic department (PBG), the University of Agriculture Peshawar, Pakistan. The procedure of Heinrichs et al. (1985) was used for screening of rice germplasm. Thirty Five (35) genotypes of rice were screened in pot experiment during 2019 including by following the method of TNI, used as susceptible check. The damage results revealed that one genotype was moderately resistant, eight genotypes were moderately susceptible, twelve genotypes found susceptible and nine genotypes were highly susceptible. The present study will help the entomologists to check the resistance factors of these varieties in detail through secondary metabolites and will help breeders in the development of leaf folder resistant varieties in future.

# STUDY ON RESISTANCE MECHANISM OF COMMERCIAL CANOLA CULTIVARS AGAINST APHID (L. ERYSIMI K. HEMIPTERA: APHIDIDAE) THROUGH ANTIBIOSIS FACTORS

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The canola aphid, L. erysimi (Kalt) is a divesting pest of canola crop of sub-content and others surrounding area of the world. Mean duration of developmental period (DP) was lower of the genotype KS-75 (3.90) followed by Oscar (4.12), while high on the Rainbow, Omega, Abaseen, Raya Anmol and Zahoor (4.90 in days). Mean reproductive period (RP) was low on variety of KS-75 (14.9) followed by Oscar (16.6 in days) while rest of the varieties were high reproductive period as compared to Oscar and KS-75. High number of fecundity (F) was produce by the variety of KS-75 (47.8) followed by the Oscar (40.2 no.) while rest of varieties were low number of fecundity produce as compared to the above two varieties. The insect pest spend more highly time longevity (L) were on the genotypes of Hoyla-401 (23.6), Omega, Dunkled, Raya Anmol and Zahoor (23.5) while low spend longevity were on the genotypes of KS-75 (18.8) Oscar and (20.7 in days) and the value of intrinsic increase (rm) of the cultivar 'KS-75' provided the antibiotic characteristics with significantly minimum intrinsic increase rate (0.605) compared to the other tested cultivars. The species spans its developmental period (DP), reproductive period (RP), fecundity (F), longevity (L) and intrinsic rate of increase (rm) in response to developmental changes of the canola plant and keeps its spreading throughout the duration of the canola crop. In generations after generation on a canola plant new born babies (nymphs) took lit a bit longer to established into adults and over the same period these old babies (adults) produced lower numbers of young ones. In the vegetative, inflorescence and fruiting stages of canola plants a higher proportion of the nymphs produced on vegetative part of the canola plant as compared to others parts of the canola plant.

# IRRADIATION OF MANGO FRUIT FLIES THROUGH ELECTRON BEAM TECHNOLOGY IN PAKISTAN

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Mango is one of the profitable fruit commodities which earn millions of dollars every year for Pakistan. Still there are many hindrances in the export of this fruit. There are many reasons behind this situation but the most problematic factor is infestation of fruit flies. In Pakistan, two species; Bactrocera zonata and B. dorsalis are most notorious. The exporters need to do disinfestations for fruit flies before exportation to meet the requirements of importing countries. Recently, e-beam technology is introduced in different countries of the world i.e., USA and China in which electron rays are used to shear the DNA of organism which ultimately kill or make them disable. In Pakistan, at port Qasim, this facility is also started to disinfest the fruits and vegetables. Therefore, this study is design to determine the effect of e-beam rays on infested Mango fruits. The experiment was laid down in complete randomized design (CRD). There were six treatments; 100 Gy, 200 Gy, 300 Gy, 400 Gy, 500 Gy and Control. Each treatment was replicated three times. In each replication, 4 Kgs of Sindhri were placed in card boxes. The mangoes were collected from the farmers' orchard and considered as infested with fruit flies. The mangoes were shifted to Pak E-beam facility port Qasim Karachi for the treatment. After treatment, the boxes were shifted to the laboratory at the faculty of crop protection for further data collection. The treated mangoes were shifted to the labeled boxes and saw dust was provided for pupation. The saw dust was sieved every week for the pupal recovery. The recovered pupae were counted and adult emergence from each treatment was recorded. The results revealed that mean number of B. zonata pupae collected from Sindhi mangoes treated with 100, 200, 300, 400 and 500 gray were 294.00±11.53, 151.33±8.97, 60.00±5.86, 33.67±4.26 and 13.67±2.33, respectively. On the other hand the mean number of recovered pupae from the untreated mangoes was 677.67 $\pm$ 14.17. There was highly significant (p > 0.01) difference was observed between all the treatments. Additionally, same trend was seen for B. dorsalis. The highest mean number of pupae were recovered from untreated fruits (355.00±8.50) followed by 100 (214.33±4.63), 200 (105.00±5.20), 300  $(31.00\pm2.31)$ , 400  $(22.33\pm2.40)$  and 500  $(10.33\pm1.45)$  Gys. Furthermore, there was no any adult emergence was observed from the treated mangoes whereas; the adult emergence of B. zonata and B. dorsalis was 89.66 and 83.76 percentage, respectively from the untreated mangoes.

# CHARACTERIZATION OF CARRIER WATER USED TO PREPARE INSECTICIDES' SOLUTION AGAINST DIFFERENT INSECT PESTS

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Currently, the failure of insecticides against different insect pests is being reported from different part of the world which ultimately results in heavy crop losses. There may be many possible reasons behind the insecticide failure but most discussed reasons are insect resistance and farmers' skills and knowledge regarding insecticide application. The most common possible reason is overlooked by the farmers' and the researchers that is quality of water used to prepare the spray tank. To characterize the underground water used for preparing tank mixture, this study was conducted under research project entitled "Characterization of Irrigation Water and Its Impact on Performance of Insecticides" funded by Higher education commission of Pakistan (HEC). For this purpose, 40 ground water samples were collected from the

tube wells and hand pumps from various locations of district Hyderabad and Tando Allahyar. The water samples were collected by using standard procedure and sent to the laboratory of Drainage and Reclamation Institute of Pakistan (DRIP), Tandojam for laboratory tests. The results revealed different qualitative parameters that can influence the insecticide performance. The ranges were; pH (6.9-8.5), Conductivity micro-S/cm (500–5240), TDS mg/1 (4.9–3,353), Alkalinity m.mol/1 (3.8–10), Hardness as CaCO3 mg/1 (60–1350), Turbidity NTU (0–93), Nitrate mg/1 (0–2.3), Carbonate mg/1 (0–30), Iron mg/1 (0.01–10.1) and Taste (unobjective-able and objective-able). Mineral constituent parameters, Bicarbonate mg/1 (190–500), Calcium mg/1 (4–220), Chloride mg/1 (18–900), Magnesium mg/1 (7.75–3159), Potassium mg/1 (1.3–14.1), Sodium mg/1 (0.3–620) and Sulfate mg/1 (8–970). The results further revealed that the pH above 8.0 can cause alkaline hydrolysis and increased dissociation (breakdown of product) and reduce the performance of insecticide formulations. Moreover, presence of calcium, magnesium bicarbonates, sulphates, chlorides and nitrates in water also can be cause of inactivation or breakdown of ions and decrease the performance of insecticide. The outcomes of this study suggest that the water should be tested prior to use for making insecticide solution and the pH and hardness must under the recommended range.

# LABORATORY BIOASSAY OF SYNTHETIC INSECTICIDES AGAINST *DYSEDERCUS CINGULATUS* (FAB.)

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Cotton stainer or Red Cotton Bug (Dysedercus cingulatus) has been recognized as one of the most destructive cotton pests in other parts of the world. It is known as the devastating insect pest of malvaceous crops including cotton, okra and sweet potato. This pest damage crops in many ways by excreting their yellowish faeces on the lint of cotton which cause to reduce the quality. The problem of resistance is a key problem in the control agriculture pests. Therefore, it is necessary to evaluate different insecticides against the key pest. In view of above facts this particular study was conducted. The laboratory experiment was carried out at Molecular Entomology Laboratory, Department of Entomology, Sindh Agriculture University Tandojam, Pakistan, during Rabi season 2018. The experiment was conducted in Complete Randomize Design (CRD). There were four insecticides includes, Dimethoate 40% EC (Dimethoate), Talstar 10% EC (Bifenthrin), Closer 240% SC (Sulfoxaflor) and Rapid 50% WDG (Acetamiprid) along with control were tested on the 3<sup>rd</sup> instar of *Dysedercus cingulatus* (Fab.). The doses were 10 ppm, 20 ppm, 30 ppm, 40 ppm and 50 ppm with different time period and data was recorded on the mortality basis after 24 hrs, 48 hrs, 72 hrs, 96 hrs and one-week. The experiment was replicated with three times, each replication contain ten 3rd instar nymphs of *Dysedercus cingulatus*. In control the nymphs were released on untreated seed of cotton crop. The overall reduction percentage in pest population was highest recorded at 10 ppm (48.1%) in Dimethoate followed by Talstar (40.7%), Closer (33.3%) and Rapid (29.6%). Similarly, at 20 ppm (59.2%) mortality was observed in Dimethoate (29.2%), whereas, minimum mortality was in Rapid (40.4%). Continuously, at 30 ppm maximum and minimum mortality was observed in Dimethoate (63.3%) and Rapid (48.1%), respectively. Furthermore, Dimethoate (70.3%) found to be highest effective at 40 ppm and 50 ppm (96.3%) during the study. In laboratory bioassay, LT<sub>50</sub> values (47.8 hrs) and LT<sub>90</sub> values (119.0 hrs) was observed in Dimethoate at 50 ppm that further confirmed, that this insecticide need short time to kill 50 to 90% population of targeted pest.

# RESPONSE OF RED COTTON BUG (*DYSDERCUS CINGULATUS*) AGAINST DIFFERENT BOTANICALS UNDER LABORATORY CONDITIONS

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Red Cotton Bug (Dysedercus cingulatus) is one the devastating cotton pests in different areas of world. This pest is mainly control by using chemical insecticides that is concern for environmentalists and public health. To replace, the botanicals are the good option. This study is conducted to screen different botanical against Red cotton bug. The Laboratory Experiment was performed at Molecular Entomology Laboratory, Department of Entomology, Sindh Agriculture University Tandojam Pakistan during Rabi season 2018. The analysis was directed in Complete Randomize Design (CRD). There were four Botanical utilized incorporates Tooh, Neem, Datura, Chilies alongside control were seen on third instar nymphs of D. cingulatus at various dosages as 12%, 14%, 16%, 18%, 20% with different time span and data was recorded on mortality premise after 24 hrs, 48 hrs, 72 hrs, 96 hrs and week. Each treatment was replicated with three replication, In every replication ten third instar nymphs of D. cingulatus were discharged on the treated seed of cotton crop. In control the nymphs were discharge on untreated seeds of Cotton crop. Be that as it may, in lab bioassay the LT<sub>50</sub>, LT<sub>90</sub>, LC<sub>50</sub>, LC<sub>90</sub> values were likewise decided against *Dysdercus* cingulatus with four unique botanicals were utilized, for example, Tooh, Neem, Datura and Chilies. The general decrease rate in pest population was most noteworthy recorded at 12% (40.7%) in Chilies comes after Neem (40.7%), Datura (37.0%) and Tooh (37.3%). Similarly at 14% (44.4%) mortality was seen in Datura pursued by Chilies (44.4%), Neem (44.44%) and Tooh (40.74%). Countiously, at 16% the most extreme mortality (51.85%) was seen in Datura comes after Neem (48.1%), Chilies' (48.1%) and Tooh (44.4%). Equivalently, at 18% (55.5%) mortality was found in Datura pursued by Neem (51.8%), Chilies (51.8%) and Tooh (48.8%). Lastly the most noteworthy population decrease rate was seen in Datura (62.9%) comes after Chilies (59.2%), Neem (55.5%) and in last Tooh (51.8%). In Laboratory Bioassy, LT<sub>50</sub> values (66.8) hrs and LT<sub>90</sub> values (186.8) hrs was recorded in Neem. Furthermore, LC<sub>50</sub> qualities (13.9) hrs and LC<sub>90</sub> values (43.1) hrs was seen in Datura at (20%) that further affirmed that Datura herbal can possibly kill 50-60% of focused population.

# INFLUENCE OF ABIOTIC FACTORS ON INSECT PESTS AND THEIR NATURAL ENEMIES ON SPRING SUNFLOWER CROP

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The study was carried out to investigate the influence of abiotic factors on insect pests and their natural enemies on spring sunflower crop. The experiment was conducted at the experimental field of Oil Seed Section, Agriculture Research Institute Tandojam, during two months and few days i.e. 29<sup>th</sup> March to 7<sup>th</sup> June. The sunflower variety HO-I were planted. The population of sunflower insect pests and their natural enemies was monitored to influence of abiotic factors on insect pests and their natural enemies on spring sunflower crop. It was observed that 24<sup>th</sup> to 31<sup>st</sup> May was the month of high jassid population (9.15±2.48 to 9.43±0.79) and the insect population trend was decreasing in 7<sup>th</sup> June (2.02±0.04). It was observed that 24<sup>th</sup> May to 7<sup>th</sup> June was the months of high whitefly population (8.22±0.48 to 14.55±0.98) and the insect population trend was decreasing in 3<sup>rd</sup> March (2.84±0.22). It was observed that 24<sup>th</sup> May to 7<sup>th</sup> June was the months of high thrips population (18.58±9.10 to 24.45±9.90) and the insect population trend was decreasing in 3<sup>rd</sup> March (5.42±1.44). The maximum mean number of *Geocoris* spp. population was recorded in the last week of March and 2<sup>nd</sup> week of April (7.00-6.67) and zero population was

recorded from 3<sup>rd</sup> week of May to first week of June. Maximum varietal mean number of *C. carnea* was recorded 1<sup>st</sup> to 3<sup>rd</sup> week of April and 1<sup>st</sup> to second week of June (1.67). There was number population of *C. carnea* from 3<sup>rd</sup> week of May to the 1<sup>st</sup> week of June. Pearson's correlation analysis between pests population and abiotic factors (temperature and humidity) and natural enemies factors is presented in (Table-5) which indicates that there was positive correlation between pests population, temperature and relative humidity indicating r-value of 0.7422 and 0.4322 respectively. But there was negative correlation between pests population and natural enemies (*Geocoris* spp. and *C. carnea*) representing r-value of -0.6983 and -0.5765 respectively. There was significant correlation between temperature and *Geocoris* spp, while non-significant for relative humidity and *C. carnea*.

### INFESTATION OF ANGOUMOIS GRAIN MOTH IN THE PRESENCE OF DIFFERENT BIOPESTICIDES AT ROOM TEMPERATURE

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The experiment was conducted in the Bio control laboratory Entomology section, Agriculture Research Institute (ARI) Tando Jam, during 2017-2018 to compare the efficacy of some plant powders against Angoumois grain moth S.cerealella (Olivier). S.cerealellais a species of gelechioid moth. It is the type species of its genus Sitotroga, placed in the subfamily Pexicopiinae of the twirler moth family (Gelechiidae). Its common name refers to Angoumois, the prerevolutionary province of France from which it was first scientifically described by G.-A. Olivier in (1789). It has been observed as a primary, serious, most harmful and commonly pest in the Angoumois province of France. It has nearly global distribution today and mainly found from tropics and warm regions. It is good flier and raises the value of infestation in wheat (Triticum sp). Wheat is a cereal grain originating from the Levant region of the Near East and Ethiopian Highlands, but now cultivated worldwide. It is grown on moreland area than any other commercial crop and is the most essential and regularly used food for humans, used to make flour for leavened, flat and steamed breads, biscuits, cookies, cakes, breakfastcereal, pasta, noodles, couscous and for fermentation to make beer (Neil 2002) and otheralcoholic beverages or biofuel. There are some different biopesticides that is prepared from leaf of various plants and make the powders. Those which were used to check the control on infestation. Were Conocarpus lancifolious (Common name: Cono), Eucalyptus citriodora (Common name: sufaido/Mushk bed), Calotropis procera (Common name: Aak) Ocimum tenuiflorum (Common name: tulsi) and Azadirachta indica (Common name: Neem). In these bio pesticides scholar checked the Mortality, Grain Weight loss and repellency by Ocimum tenuiflorum (Common name: tulsi) by following with CRD with four replica. The level of infestation is shown by all five extracts. The level of similarly effectivenesswas utilized in preventing grain weight loss was also observed. The most effectiveness in case of preventing the grain damage and extracts of plants were toxic to adult moths and also prevent hatching of the eggs of S. cerealella. It was showed by Ocimum tenuiflorum (Common name: tulsi) in all rest of others It was observed that Ocimum tenuiflorum(Common name: tulsi) is most effective. Egg incubation period of Angoumois grain moth was (4.75 days), The duration of 1<sup>st</sup>, 2<sup>nd</sup>, 3<sup>rd</sup>, 4<sup>th</sup> and 5<sup>th</sup> instar was (8.75, 12, 15.5, 18.5 and 22.25 days), respectively. The pupal development period of Angoumois grain moth was (13.00 days), Adult development period of Angoumois grain moth was (16.66 days).

### PROTECTION OF FRUIT ORCHARDS FROM RAVAGES OF FRUGIVOROUS BATS IN PAKISTAN

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Fruit bats, *Pteropus giganteus*, commonly known as flying fox belong to order *Chiroptera*, class *Mammalia*, is found all over South Asia, from Bangladesh to India, Nepal and Sri Lanka. In Pakistan, fruit bats are found

throughout the country from KPK, Punjab, Sindh and Azad Jammu & Kashmir. They provide substantial ecological and economic services via pollination, seed dispersal and are categorized as agricultural pests. Fruit bats, being flying mammals, have excellent sense of smell, gregarious nature and feed on un ripe and ripe fruits. They are reported to attack apple orchards in upper Swat, feast on lychees in Haripur and feed on mangoes and dates in Sukkur and Mirpur Khas districts of Sindh. In Khanpur area, Haripur KPK, farmers were deprived of up to Rs 0.4 million losses to lychee crop in a single season. Furthermore, fruit bats are reported to be possible natural reservoirs for lethal Ebola virus strains in addition to being carrier and reservoir of Hendra and Nipha viruses. These are fatal in human and other mammals as cases reported in Australia, Malaysia, Singapore, India and Bangladesh. Among possible causes of diversion of fruit bats to fruit orchards damage are high rate of deforestation for cultivation of land, degradation of natural habitats and rapid urbanization. Management strategies for fruit bats include use of mist nets, scaring devices application and reforestation for their roosting and food requirements. Arrangement of awareness campaign through seminars, workshops and electronic media is recommended. Detail in-depth study is suggested to ascertain biodiversity, effect of climate changes and effectiveness of different management techniques to save fruit orchards from their attacks.

#### IMPACT OF PB ROPES ON POPULATION DYNAMICS OF JASSID, THRIPS AND THEIR NATURAL ENEMIES IN COTTON

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Cotton is most important fiber crop of Pakistan. Cotton contributes 1.6% in GDP of Pakistan. Jassid (Amrasca biguttula biguttula) and thrips (Thrips tabaci) are major sucking insect pests of cotton. They are serious threat to many agricultural crops including cotton. This research focused on ecological impacts of PB ropes on population dynamics of sucking insect pests of cotton (jassid and thrips) as well as beneficial insects. Research was conducted at MNSUAM Experimental Farm C-block on an area of 27 acres near old Shujabad road Multan. The research field was divided into 6 blocks. Each block consisted on 5 acres. Cotton variety NIAB 878 was cultivated as test crop. Population data was record on weekly base from twenty random plants from each acre for sucking insect pests and ten plants for beneficial insects. Statistical analyses were done by following ANOVA and SEM was separated using statistics 8.1. The results revealed that population of jassid and thrips remained significantly lower in treatment T2 (PB-ropes+PB-ropes) as compared with treatment T1 (PB-ropes+insecticides), while maximum population was denoted in control block which was managed through use of pesticides. Key insects (predators) suppressing the population of jassid were spiders and campylomma, they showed negative correlation against jassid hence played role in reduction of infestation. While in case of thrips all recorded beneficial (green lacewing, orius, geocoris, campylomma and spiders) showed negative correlation and kept its population at minimum level as compared with control.

# CONTROL OF WELL-KNOWN QUARANTINE PEST 'SPODOPTERA FRUGIPERDA' (FALL ARMYWORM) ON MAIZE CROP THROOUGH UV LIGHT TRAPS

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The genus "Spodoptera" has many species but *Spodoptera frugiperda* is economically important species for agricultural crops worldwide. It is mostly found in tropical and subtropical regions. *S. frugiperda* belongs to Family "Noctuidae" and Order "Lepidoptera. *S. frugiperda* (Fall Armyworm) is an invasive pest of many crops but it is a most serious pest for maize crop. It is migratory lepidopteran insect pest with more than 100 host plant species. *S. frugiperda* belongs to Family "Noctuidae" and Order "Lepidoptera". Its larval stages feed on the leaves of the maize plants. When population increases it starts to feed on the ears of the maize plant. S. frugiperda can be controlled by various types of

insecticides worldwide but now it starts to develop resistance against many insecticides. So present were designed to evaluate the effectiveness of UV light traps in maize crop, at Maize and Millet Research Institute, Sahiwal during 2019. Five UV light traps were installed in 6 acres of maize crop. Result revealed that UV light traps reduce 60% population of *S. frugiperda*. So by this control method we can save our ecosystem and nature from hazardous effects of chemicals.

### IMPACT OF HEAT STRESS ON THE BEHAVIOR ADOPTABILITY OF RICE LEAF FOLDER (CNAPHALOCROCIS MEDINALIS) UNDER LAB CONDITION

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The whole world is facing a very serious issue now a days known as a climate change and its increasing day by day in a very speedy way. The overall atmospheric temperature has jumped up to 0.7°C in last 100 years. Global warming is affecting the all the organisms and insects are one of them. Rice is very important crop in the world and attacked by an important insect pest, Rice Leaf Folder (*Cnaphalocrocis medinalis*). Climate change has reduced the capability of rice leaf folder to fold the leaf hence reducing the survival rate. Therefore, the behaviors of larval and adult rice leaf folders, such as leaf folding (making shelter) and habitat selection for pupae and eggs, were observed on rice plants under heat stress at Integrated Genomic, Cellular, Developmental and Biotechnology Laboratory, University of Agriculture, Faisalabad. The results showed that heat stress for four hours decreased the shelter making power and shelter size also reduced. Larvae favored to pupate on young rice leaves, meddle-aged leaves and older leaves at 25°C, 28°C and 33°C, respectively. Normally, female moths lay eggs on top young leaves but they deposited eggs on middle and lower leaves on exposure to temperature 35°C and 38°C, respectively. Moreover, on increasing heat stress more eggs were deposited on lower surface of leaves. The study concluded that rice leaf folder can adopt heat stress, so there will be high chances of outbreak under global warming.

### BIOLOGICAL PARAMETERS OF *TRILOCHA VARIANS* (BOMBYCIDAE, LEPIDOPTERA), A PEST OF *FICUS BENJAMINA*

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Ficus benjamina is commonly known as weeping fig, planted alongside the road in various regions of the world including Pakistan. Weeping fig is native to tropical and subtropical regions of the globe. It increases the aesthetic value of the country (Pakistan). Trilocha varians (Bombycidae: Lepidoptera) is the major insect pest of Ficus benjamina. It has close relationship with domesticated silkworm, Bombyx mori. There is need to determine the biological parameters of this pest. For this purposes, the current study was conducted in rearing laboratory at Institute of Plant protection, MNS-University of Agriculture, Multan during April-June 2019 on Ficus benjamina and Ficus virens. The study revealed that fecundity of female was range from 164 to 275 which increased its survival rate. There were five larval instars. The fourth and fifth instars were dark reddish. A caudal horn was present on each larva. The larva of pest was caused 100% defoliation and even killed the whole Ficus plants. Larvae feed vigorously and can consume all of the leaves of an individual shrub in just overnight. The male was fast flayer and long lived than female. The information about biological parameters of Trilocha varians contained in this paper will proved suitable management strategy control this pest outbreak.

### EFFICACY OF ENTOMOPATHOGENIC FUNGUS, BEAUVERIA BASSIANA AGAINST DIAPHORINA CITRI UNDER FIELD CONDITIONS

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Diaphorina citri is a key pest of citrus (rutaceae). It is considered a vector of phloem feeding bacterium, Candidates liberibacter resulting citrus greening disease throughout the globe. Entomopathogenic fungi are potential candidate and most suitable alternative strategy to control D. citri. An experimental study was conducted to check the efficacy of Beaveria bassiana against D. citri in Multan 2019. For this purpose, commercial formulation was imported from China and different doses of fungus tested against this pest by using Randomized Complete Block Design (RCBD). The mortality data were recorded at 4<sup>th</sup> and 7<sup>th</sup> days of post treatment. The study resulted that the tested entomopathogenic fungus was cause 70.9% mortality. The mortality rate was increase with increase in dose and time. The population of D. citri was reduced at 7<sup>th</sup> day as compared to 4<sup>th</sup> day. During the study, no mortality was observed in control treatment. The current study concluded that tested fungus is an effective tool to control D. citri under laboratory as well as field conditions in country.

### EFFICACY OF BIO-LARIVICIDES AGAINST DENGUE VECTORS; AEDES AEGYPTI & AEDES ALBOPICTUS UNDER LABORATORY AND FILED CONDITIONS.

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Aedes aegypti & Aedes albopictus act as the key vectors for many human diseases such as dengue fever, dengue hemorrhagic fever, chikungunya etc. causing millions of deaths every year. Due to the insecticidal contamination of the environment and behavioral resistance in vectors, increasing attention is now being focused on environmental friendly control methods. The use of easily degradable plant based compounds are considered to be one of safest, cheapest approach of insect pests control especially dengue vectors as an alternative source for the synthetic chemicals. Therefore, the present study was made to monitor the larvicidal properties of various different plant extracts against 3rd & 4th instars larvae of two Aedes species. These extracts were applied at three different dosage rates i.e., 1, 2 and 3% and 1-2 days of exposure time under laboratory conditions. Results showed that the parthenium & black pepper were much effective (100% mortality) at 24 hours exposure period and 1% concentration. Similarly garlic has shown the mortality effect as 56.66% at 24 hours exposure time and 3% concentrations. When the dosage rate and exposure time of the plant extract was increased, the mortality rate was also amplified. After 48 hours of exposure time, the garlic has shown significant mortality effect of (80-90%) at 3% concentrations. Similar mortality trend was observed in mint extracts. While even at the highest dosage rate i.e 3% of onion extract showed the minimum mortality as (13.33%) after 48 hours exposure time. In the control treatment only (3%) mortality was found at 48 hours of exposure. The efficacy in term of mortality were found in order of Parthnium and black pepper > garlic > mint >onion. Thus environment friendly plant based pesticides can be integrated with other available techniques for population suppression of larvae and can be successfully included in the dengue vector control program.

# EFFECTIVE MANAGEMENT OF THE LESSER BANDICOOT RAT (BANDICOTA BENGALENSIS) IN WHEAT IRRIGATED CROPLAND ECOSYSTEM WITH TRAP-BARRIER SYSTEM

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Predominantly, impacts of rodents on both the stability and sustainability of productive agro-ecosystems cannot be denied. Invariably, they remain potential destructive agents of crops, stored grain structure, indoor situations, grain markets and residential plots. Therefore, their depredations and economic pillage almost always leads to intensive economic losses. Incidentally, wheat (*Triticum aestivum*) remains one of the important food crops of South-East Asia and is frequently predicated. Due to the intensive damage pattern of rodent pests, there is always decline in its annual production. Therefore, present situation demands ecologically acceptable strategy which can delimit the rodent pest elimination from various situations. In central Punjab, most prominent vertebrate pest was the lesser bandicoot rat (*Bandicota bengalensis*) which specifically focuses on the agriculturally important crops including that of the wheat. Conclusively, it was evident that Trap barrier system (TBS) was an innovative method which can successfully delimit the impact of rodent infestations by ecologically management of the recyclable poly-ethylene sheets inserted into the wheat crop along with the placement of varied traps viz. Snap tarp, multi-catch trap, glue station and glue trap incorporated with suitable baits for efficient rodents captured on weekly duration. The data was statistically analyzed using a personal computer with analysis of variance, complete randomized designed and correlation statistical analysis to describe the result of rodent captured.

## PREFERENCE OF PUPAL PARASITOID *DIRHINUS GIFFARDII* ON *BACTROCERA ZONATA* PUPAE REARED ON DIFFERENT DIETS IN LABORATORY.

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The peach fruit fly *Bactrocera zonata* (saund) is recognized as one of the most significant and well established serious pest of fruit trees of south and south east Asia. *Bactrocera* spp. are considered as serious pests of fruits in Pakistan. The research was conducted at the Dipterian Research Laboratory Department of Entomology, Sindh Agriculture University Tandojam. Five fruit host viz Banana (Musa Cavendish), Guava (Psidium guajava), Chikoo (Manilkar azapota), Orange (Citrus aurantium) and Ber (Ziziphous jujube), five hundred gram each were placed in cage for the oviposition of Peach fruit fly *Bactrocera zonata* for two hours. After two hours the host fruits were separated in five different cages containing fine saw dust at the bottom for pupation. The results of research study revealed that maximum male and female emergence of *B. zonata* observed on guava fruit followed by banana, Ber, Chikoo and Orange respectively. The preference of pupal parasitoids *Dirhinus giffardii* was observed for 48 hours at different ages of *B. zonata* pupae, i-e 24, 48, 72 and 96 hours for parasitism. After 48 hours *B. zonata* pupae kept into different jars to observe the parasitism. The result indicated that minimum parasitism was recorded in 24 hours old pupae and maximum parasitism was recorded in 72 hours old pupae. The impact of different photoperiods for parasitism of *Dirhinu giffardii*, *i.e* T<sub>1</sub> = 8/16, T<sub>2</sub> = 10/14, T<sub>3</sub> = 24/0, T<sub>4</sub> = 0/24 (L/D = Light / Darkness) were observed. The maximum parasitism was observed at photoperiod (24/0 L/D) and minimum (0/24L/D).

### SEASONAL MONITORING OF *BACTROCERA* SPP. USING METHYL EUGENOL TRAPS IN GUAVA ORCHARD HYDERABAD-SINDH

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Bactrocera species are serious threat to production and quality of fruits particularly mango and guava in relation to Sindh Province. Male adult population of fruit fly species captured through methyl eugenol pheromone traps installed of different heights *i.e.*, Surface, 1 metre, 2 metres and 3 metres. All were installed at Nuclear Institute of Agriculture (NIA) guava orchard, Tandojam District Hyderabad with 1340.56±24.92, 1185.38±15.66, 1428.36±20.11 and 1177.32±24.24 trap catches while B. dorsalis catches in methyl eugenol pheromone traps with 6.29±0.02, 4.95±0.02, 7.35±0.03 and 4.67±0.02 trap catches. Regardless the trap height B.zonata and B. dorsalis infestation was highest when the methyl eugenol pheromone traps were installed at 2 metre height; while traps installed at 1 metre and 3 metres height could not catch B. zonata and B. dorsalis more than surface installed traps.

### DISINFESTATION OF CHICKPEA (CICER ARIETINUM L.) FROM CALLOSOBRUCHUS CHINENSIS ADULTS BY USING MICROWAVE OVEN

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Chickpea (*Cicer arietinum* L.) is an important cash crop of Pakistan. During storage *Callosobruchus chinensis* (Coleoptera: Bruchidae), causes severe post-harvest losses. In the present study, we aimed to test the mortality of *C. chinensis* adults by using different powers of microwave oven under laboratory conditions. Healthy chickpea *was artificially infested with C. chinensis* and then heated in microwave ovens (2,450 MHz) of different power outputs at 1000, 1100, and 1200 W for 0, 15, 30, 45, and 60 s. Germination test was also performed to test the germination ability of chickpea after exposure to microwave radiations. The results revealed complete mortality of *C. chinensis* after 30, 45 and 60 s at all powers but the change in colour of commodity was observed after 45 and 60 s. However, germination ability was not effected at all concentrations and exposure timings. The study suggested that microwave irradiation can be used as an appropriate alternative method for management of C. *chinensis* and other pests of stored grains.

# INCIDENCE OF INSECT PESTS ON BER ZIZIPHUS JUJUBE AND THEIR MANAGEMENT THROUGH INTEGRATED PEST MANAGEMENT TECHNIQUES

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The studies on incidence of insect pests and their management Integrated Pest Management techniques was conducted at PARC-Arid Zone Research Institute, Umerkot, during the season 2018-2019. Experiments were comprised on the insect pest population in jujube tree, performance of pheromone traps at different heights for mass trapping of fruit fly adult, Monitoring the population of natural enemies of fruit fly, The comparative effectiveness of

different pesticides against major sucking pests was conducted in AZRI field. The results revealed that insect pests as termites, fruit fly, leaf hopper, leaf miner, bark beetle and grey weevil was found on ber plants, Whereas, maximum population was observed termites as compared to other insects. A maximum fruit fly and termites population was observed in the month of January. The maximum activity of grey weevil and leaf miner was found in the month of May. Highest population of bark beetle and leaf hopper was observed in the month April The results regarding male adult collection at different heights there was no significantly different between different heights. The result revealed that the maximum population 12.50±1.05 recorded at 4 feet height, followed by at 6 feet height 12.14±1.05, and at 2 feet 11.56±0.30. The results revealed that 2 feet height was less effective for mass trapping of fruit fly during the season. The use of pesticides on jujube field was found effective against leaf hopper. Bifenthrine insecticide was more effective against leaf hopper and Emamectine was found highly toxic against leaf miner other insecticides.

### POPULATION DYNAMIC OF LEAF HOPPER ON BRINJAL WITH RELATION TO WEATHER PARAMETERS IN MULTAN

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An experimental study was conducted to check the population dynamics of leaf hopper on brinjal in relation to weather parameters under field conditions at farmer field during 2019. The leaf hopper population was started after crop plantation. The highest population of leaf hopper was recorded from 21<sup>st</sup> May to 7<sup>th</sup> August. The highest number of leaf hopper per plant was noticed as 9.7. The pest population was highly affected with weather parameters such as temperature. The maximum and minimum temperature were found as positively and significantly corrected while relative humidity (RH) was found negatively and non-significantly correlated with pest population change. The sunshine was also found positively and non-significant correlated factor.

# ALTERNATE HOST PREFERENCE OF *BACTROCERA SPP*. AND HOST SUITABILITY OF DIFFERENT PUPAL AGE OF *BACTROCERA* SPP. FOR THE PARASITOID, *PACHYCROPOIDEUS VINDEMMIAE* UNDER LABORATORY CONDITIONS

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Fruit fly (Diptera: Tephritidae) are most important pest in different regions (tropical, sub-tropical and several temperate) in the world. They attain international importance due to cosmopolitan nature in fruits and vegetables production and trade issues. Current experiment was designed to evaluate host preference of three *Bactrocera* species for oviposition and parasitism potential of *P. vindemmiae* under lab condition at Integrated Pest Management Lab, University of Agriculture, Faisalabad. Fruit fly species were collected from the different field from district Faisalabad and Sargodha. These species were reared on its natural host as well as artificial host under control condition. Identified species were released in cages to check host preference for oviposition. Result showed that guava, Ak fruit and pumpkin were the most preferred for all Bactrocera species. Parasitism of *P. vindemmiae* on pupae of *B. dorsalis*, *B. cucurbitae* and *B. zonata* was showed 25%, 37%, and 38%. Statistically analysis showed that the rate of parasitism of *P. vindemmiae* was maximum for pupae *B. zonata* and *B. cucurbitae*. Parasitism potential of parasitoid was maximum on 2-4 days old pupae. So, it can be concluded that the pupal parasitoids *P. vindemmiae* can be effective biocontrol agent for integration in pest management of fruit flies.

### EVALUATION OF *TRICHODERMA HARZIANUM* AND *AZADIRACHTA INDICA* IN THE MANAGEMENT OF *MELOIDOGYNE INCOGNITA* IN TOMATO

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Root-knot nematodes of the genus *Meloidogyne* badly affect crop production worldwide and cause huge yield losses. Among the known root-knot species, Meloidogyne incognita is by far the most destructive, widely distributed and the most dominant and prevalent. In the present study, the efficacy of a biological control agent, Trichoderma harzianum and an antagonistic plant, Azadirachta indica was tested against M. incognita on tomato. The antagonistic fungus and plant caused significant hatching inhibition and larval mortality of M. incognita. The hatching inhibition and mortality was the maximum at 100% concentrations of both the agents while the minimum inhibition and mortality was obtained at 25% concentration. No statistical difference was observed between T. harzianum and A. indica in causing hatching inhibition and larval mortality. Numbers of galls, egg masses and reproductive factor were reduced significantly as a result of A. indica and T. harzianum applications. The reductions in these parameters were more pronounced where both the agents were integrated and resulted to the maximum where both the agents were mixed at the highest concentrations. The reductions in galls, egg masses and reproductive factor were found inversely proportional to the concentrations of A. indica and T. harzianum. Similarly, all the concentrations of A. indica when integrated with T. harzianum increased plant height and fresh shoot weight significantly over control. The increase in these parameters was directly proportional to the concentrations of A. indica leaves and T. harzianum. A. indica amendments proved at par with those of T. harzianum. The maximum increases in these parameters were obtained where A. indica leaves were mixed at the rate of 50 g with 6 g of T. harzianum. The amendments also showed significant effects on root weight. The maximum decrease in root weight was observed where both A. indica and T. harzianum were mixed at the highest concentrations. The root weight decreased with an increase in concentration and was found inversely proportional. It is therefore, recommended that the integration of antagonistic plants with the antagonistic fungi may be useful for the better control of plant parasitic nematodes.

#### IMPACT OF SPICES ESSENCES WITH METHYL EUGENOL TO ATTRACT BACTROCERA SPP. AT GUAVA ORCHARD

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The fruit flies are the most key pests of many soft fruits over all the world. Fruit flies found in tropical Asia and has been spread in several tropical countries of Asia. *Bactrocera* spp. are serious threat to production and quality of fruits particularly Guava, Chiku, Mango and Jujube in relation to Sindh Province. Male adult population of fruit fly species captured through spices essences were tested with Methyl eugenol i.e. Nutmeg essence (T1), Fennel essence (T2), Cardamom essence (T3) and control Methyl eugenol (without any essence) (T4). All were installed at Guava orchard, Agriculture Research Institute (ARI) Tandojam. Regardless the traps *Bactrocera* spp. infestation was highest at Nutmeg essence with methyl eugenol pheromone traps; while traps installed at Cardamom essence, control Methyl eugenol (without any essence) could not catch *B.zonata* and *B.dorsalis* more than Fennel essence.

### INFESTATION OF RED FLOUR BEETLE (*TRIBOLIUM CASTANEUM*) ON DIFFERENT NUTS UNDER LABORATORY CONDITION

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The laboratory experiment was conducted on the infestation of red flour beetle (Tribolium castaneum) on four different nuts like Pine nuts, Walnuts, Cashew nuts and Peanuts under laboratory temperature was 28±2°C and relative humidity was 75±5% in the Department of Plant Protection, Faculty of Crop Protection, Sindh Agriculture University, Tandojam during 2018. For that particular research study 50g of each sample of different nuts were used for observation. According to the results 145.00±7.64 maximum mean population of red flour beetle was observed on 20th July, 2018 and 14.00±1.00 was minimum mean population was recorded on 5th April, 2018 in Pine nuts. Similarly, the highest mean population of red flour beetle was reached at 18.33±0.67 on 21st March, 2018 and 1.67±0.33 was lowest mean population observed on 20th June, 2018 in the Walnuts treatment. The data further revealed that the population of T. castaneum was more observed in Cashew nuts on 20th July, 2018 mean population was 37.00±11.93 followed by 8.33±1.76 on 5th June, 2018. According to the last treatment, the peak mean population was observed i.e. 13.67±8.41 on 20th July, 2018 whereas, 6.00±2.00 lowest mean population was recorded on 20th April, 2018 in Peanuts. While 80.11±16.91 overall maximum mean population was observed in Pine nuts followed by Cashew nuts (15.78±2.86), Peanuts (9.81±0.77) and Walnuts (6.96±1.92), respectively. According to the weight loss data, the highest percentage of weight loss of Walnuts was recorded 0.15% and the lowest weight loss percentage was observed 0.02% in remaining nuts like Pine nuts, Cashew nuts and Peanuts. Similarly, 7.56g maximum weight loss of T. castaneum was recorded in walnuts followed by Pine nuts (6.09g). While 1.00g minimum weight loss was recorded in cashew nuts and peanuts. The data analysis a significant different (P < 0.01) in the overall population of *T. castaneum* among various treatments.

### EFFICACY OF NEW CHEMISTRY INSECTICIDES AGAINST BACTROCERA ZONATA (DIPTERA: TEPHRITIDEA) AND THEIR BEHAVIOR IN CITRUS FRUITS

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Direct and indirect damage caused by fruit fly to agricultural fruit crop and vegetables throughout the world. Fruit fly is quarantine pest so infested fruit and vegetables restricted to export. Citrus fruit is important fruit crop in Pakistan. Fruit fly have high fecundity, short life cycle and wide host ranges that why difficult to control. Against *Bactrocera zonata* the current study was conducted to check efficacy of new chemistry insecticides and their behavior in citrus fruits. Feeding behavior was determined the twenty mixed gender fruit flies was released into each dish. Count the numbers of fruit fly which was feed on treated and control filter paper. The data was collected and analyzed using suitable statistical techniques. The study was useful for the future management of fruit fly in citrus. Results showed that bottle dip method showed that the highest mortality was recorded in maximum concentration of trichlorfon at 2 ppm (100 %) after 72 and lowest mortality was found in control treatment after 24, 48 and 72 hours (6 %), (6.8 %) and (8.6 %) respectively. The results pertaining the feeding behavior showed that trichlorfon the fruit fly was settle on treated plant after 24 hours (7), 48 hours (8) and 72 hours (8). The novluron treated plant showed that after 24 hours (11), 48 hours (7) and 72 hours (8). Hence it was concluded that for the control of fruit fly trichlorfon best by using bottle dip method and for feeding behavior fruit fly also prevent on the tree or plant which was spray by insecticides. This study can play important role for the management of fruit fly in Pakistan.

### RESPONSE OF BACTROCERACUCURBITAE TO CUE-LURES IN VARIABLE TRAP SHAPES IN BITTER GOURD

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Bitter gourd (*Momordica charantia* L.) is most cultivated crop in Asian region and it has remarkable value because of its nutritional and medicinal value. Bitter gourd quality and production decreases by *Bactroceracucurbitae*. Fruits affected and contaminated by fruit fly are unfit for human consumptions. *B.cucurbitae* was not attracted by traditional lures used for other species of *B. cucurbitae*. For this purpose, methyl eugenol and Cue-lure was applied in farmer field Multan for trapping and monitoring of *B.cucurbitae*. Experiments were performed in 4 acre of bitter gourd field and plots were divided in four parts A, B, C and D. To observe the efficiency of traps, plots A and B were treated with methyl eugenol and *B.cucurbitae*lure, plot C and D was remained as untreated plots. Two different shapes of trap (bottle and cylinder) were also used for attraction and observed the efficiency of lure against target insect. Results depicted that maximum fruit flies were captured in cylinder shape trap baited with *B. cucurbitae* lure trap. However, weight of healthy fruits and damaged fruits was also recorded for comparison. It was observed that production increased in treated plots as compared to untreated plots.

### FIELD EFFICACY OF ENTOMOPATHOGENIC FUNGI FOR MANAGEMENT OF LEUCINODES ORBONALIS

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Eggplant (Solanum melongena L.) is an important vegetable crop throughout the world particularly in tropical and subtropical regions. Different insect pests attack on it from the date of sowing to harvesting and Leucinodes orbonalis (Guenee) is one of the most damaging insect pest. A single borer can infest 4 to 7 fruits and various shoots of plant during its lifecycle. Synthetic pesticides are preferred choice of farmers irrespective for hazard to human health and environment. Excessive use of insecticidess makes eggplant fruit as a big source of toxic residues. Under these circumstance, eco-safe agents such as entomopathogenic fungi (EPF) are best alternate for its management. An experiment was conducted to evaluate the efficacy of EPF against L. orbonalis. For this purpose Beauveria bassiana and Metarhizium anisopliae were evaluated under different treatments which are [B. bassiana @ 2.5 g/L and 3.5 g/L, M. anisopliae @ 2.5 g/L and 3.5 g/L]. Data was recorded 1st, 3rd, and 7th day of treatments. Our results depicted that B. bassiana provided highest moratlity at 3.5 g/L concentration as compared to other treatments. Hence, B. bassiana can be recommended as a bio-control agent in integrated pest management component on eggplant because of effective and environmentally friendly.

### DISTRIBUTION OF COMMENSAL RODENTS AND PREVALENCE OF HELMINTHIC INFECTION IN THREE DISTRICTS OF HYDERABAD DIVISION, SINDH, PAKISTAN

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The aim of present study was to investigate the distribution of commensal rodents and their helminthic infection in Hyderabad division. During this research work three rodent species *Mus musculus* (House mouse), *Rattus rattus* 

(Back rat) and Rattus norvegicus (Brown rat) were collected from different structures (home, shop and street) of three districts including Hyderabad, Matiari and Jamshoro during the months of May to October, 2019. A total of 52 samples were collected. In district Hyderabad only Rattus rattus (Back rat) was captured with relative estimates of abundance (59.7%), whereas, no Mus musculus (House mouse) and Rattus norvegicus (Brown rat) were collected. In district Jamshoro the Mus musculus (House mouse) was captured with relative estimates of abundance (77.6%) and Rattus rattus (Back rat) with (56.91%), whereas, no Rattus norvegicus (Brown rat) were collected. In district Matiari the Mus musculus (House mouse) was captured with relative estimates of abundance (75.2%), Rattus rattus (Back rat) with (40.4%) and Rattus norvegicus (Brown rat) with (23.3%). In helminths, the maximum prevalence of helminths was found in district Jamshoro (61.10%). In helminths the maximum prevalence (70%) was recorded for nematodes followed by cestodes (37.5%). None of the trematode and acathocephala was recovered from hosts of this district. In district Hyderabad the prevalence of helminths was recorded (36.5%). Among them, maximum prevalence (70.3 %) was recorded for cestodes, followed by nematode (28.9%). The minimum prevalence (0.78%) was recorded for trematodes. None of the acanthocephalan was collected from hosts of this district. In districts Matiari, the prevalence of helminths was recorded (9.2%). Among helminths maximum prevalence (88%) was recorded for the nematodes followed by cestodes (12%). None of the trematode and acathocephala was recovered from hosts of this district. The collected data were analysed statistically by using Chi square statistical procedures. Present study is the part of research project no. 9412/Sindh/NRPU/R&D/HEC/2017.

### INTEGRATION OF SEX PHEROMONES AND PESTICIDES FOR THE MANAGEMENT OF SPOTTED BOLLWORM (LEPIDOPTERA: NOCTUIDAE)

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Spotted bollworms Earias insulana (Boisduval) and Earias vitella (Fabricius) have become most destructive and severe pest of cotton and okra in Pakistan especially the last few years. The pest caused serious losses despite the regular use of pesticides. In the current study, the integration of sex pheromones and pesticides for the management of spotted bollworm carried out in Ayub agricultural research station, Faisalabad. To monitor the adult population, the sex pheromones Z11- Hexadecenal Z11 octodecenal (10:2:2) and (10E 12E) – 10, 12 Hexadecadienal were installed for Earias vitella and Earias insulana. The traps were inspected once a week till the harvesting of the crop. The lures/capsules were replaced after every 15 days to keep capturing effective. The collected data was subjected to Tukev's test for data analysis. The results showed that total 1067 adults of Earias vitella and 458 Earias insulana were caught during whole cropping season in six traps. The maximum capturing (38) and (34) were recorded during mid of September 2018 in two cotton varieties. On the other hand, minimum moth catches (2) and (3) were noticed in end of November of two cotton varieties respectively. The results also showed that maximum boll damage percentage 10.16 and 9.5% was noticed in non-Bt (FH942) cotton variety in last week of September which is followed by 1st week of October in cotton crop respectively. This was comparatively high from in Bt (FH-142) in the end of September which is followed by start of October in cotton varieties. On the other hand, minimum damage percentage (5.83%), (4.83%) in non-Bt (FH-942) was monitored in start of November. The maximum damage (8.33%), (8.16) of flowers was noticed in non-Bt (FH-942) and (3.33%), (3.16) in Bt cotton varieties in last two weeks of September. On the other hand, minimum damage percentage of bolls were (5.83%), (4.83%) in the first two weeks of November and the minimum damage percentage of flowers were (1.33%), (1.66) in start of July in non-Bt (FH-942). The result also concluded that Bt (FH-142) has maximum yield (540 kg) and (521 kg) in treated and untreated cotton crop respectively. These are comparatively high from non- Bt (FH-942) which has minimum yield (470 kg) and (455 kg) in treated and untreated cotton varieties. The results showed that sex pheromones can be integral part of IPM strategies for spotted bollworm species.

### EVALUATION OF ENTOMOPATHOGENIC FUNGI AGAINST PINK BOLLWORM OF COTTON PECTINOPHORA GOSSYPIELLA SAUNDERS

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In Pakistan, pink bollworm recognized as the major insect pest of cotton that responsible for significant yield losses. It not only reduces the quantity of the crop through direct feeding, but their existence may also damage the lint quality of cotton. The yearly losses by this pest in Pakistan are estimated about one million bales. Several methods are in use for killing or preventing insect pest issues, but in most cases the remedy to insect pest infestation is usually obtained by the use of chemical insecticides. However, the frequent applications of synthetic insecticides caused many disadvantages such as elimination of natural enemies, development of insecticide resistance and accumulation of toxic substances in the environment. Therefore, in present study, we tested entomopathogenic fungi including Metarhizium anisopliae, Aspergillus niger, Paecilomyces lilacinus, Trichoderma polysporum, Aspergillus flavus, Fusarium solani, Beauveria bassiana and Paecilomyces variotii against Pectinophora gossypiella. The larval mortality of pink bollworm varied greatly with the entomopathogenic and their concentration applied. Among eight fungi tested against larvae of Pectinophora gossypiella, the most effective were Metarhizium anisopliae and Aspergillus niger followed by Paecilomyces lilacinus. The medium and higher dose of Metarhizium anisopliae and Aspergillus niger, and higher dose of Paecilomyces lilacinus caused mortality of all larvae of pink bollworm. The highest concentration of Trichoderma polysporum and Aspergillus flavus was moderately effective; while Fusarium solani, Beauveria bassiana and Paecilomyces variotii appeared less effective or ineffective against the targeted pest. The maximum pupation took place in Fusarium solani followed by Paecilomyces variotii and Beauveria bassiana. No or very less pupation occurred in larvae treated with Metarhizium anisopliae and Aspergillus niger, followed by Paecilomyces lilacinus. In most treatments, pupation was decreased with increasing concentrations of entomopathogenic fungi. No adult emergence was observed in all three concentrations of Paecilomyces lilacinus, Metarhizium anisopliae and Aspergillus niger. On the other hand, the maximum adult emergence of pink bollworm was noted in different concentrations of Fusarium solani and Beauveria bassiana ranging from 60 to 90.9%. The second highest adult emergence was recorded in different concentrations of Paecilomyces variotii, Aspergillus flavus and Trichoderma polysporum ranging from 40 to 71.4%. Further studies should be carried out to test the efficacies of entomopathogenic fungi against pink boll worm and other insect pests under field conditions.

# LABORATORY EVALUATION OF ENTOMOPATHOGENIC FUNGUS *BEAUVARIA BASSAIANA* AND *METARHIZIUM ANISOPLIAE* AGAINST 2<sup>ND</sup> AND 4<sup>TH</sup> INSTAR LARVAE OF RED PALM WEEVIL *RHYNCHOPHORUS FERRUGINEUS*

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The date palm (*Phoenix dactylifera* L.) belongs to family Arecaceae. It is considered a sign of life in deserts, because as compare to other fruit crops it stands in extreme temperatures survive in drought and salinity conditions. The Red Palm Weevil *Rhynchophorus ferrugineus* is a globally obnoxious insect pest of date palm (*Phoenix dactylifera* L.). The *R. ferrugineus* generally referred as concealed tissues borer; complete its life cycle inside the palm tree Different pesticides are widely used for the management of RPW. Used of pesticides causes different problem such environmental pollution, development of resistance and adverse effect on non-target species. In order to reduce the use of insecticide, the application of *Beauveria bassiana* and *Metarhizium anisoplae* may be an alternative control. The Red Palm Weevil *Rhynchophorus ferrugineus* is a globally obnoxious insect pest of date

palm (*Phoenix dactylifera* L.). Different concentrations of *Beauveria bassiana* and *Metarhizium anisoplae* were applied against 2<sup>nd</sup> and 4<sup>th</sup> instar larvae of *R. ferrugineus*. Larvae of 2<sup>nd</sup> and 4<sup>th</sup> instar were dipped for 60 s in fungal suspension. Data of mortality was recorded after 7, 14 and 21 days. The results of present study shows that 2nd instar larvae of RPW are more susceptible as compare to 4th instar larvae. Fungal spore penetrates easily in cuticle of 2nd instar easily because its body is more delicate and soft as compare to 4th instar. 100% mortality of 2<sup>nd</sup> instar larvae was recorded in treatment Ma-3 after 21 days of application. Less mortality observed in treatment Bb-1 after7 day application.

### EFFICIENCY OF PHEROMONAL AND ELECTRIC TRAPS AGAINST *BACTOCERA ZONATA* (TEPHRITIDAE: DIPTERA) IN MANGO ORCHARDS AT DISTRICT KHANEWAL

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Mango (Mangifera indica) is the national fruit of Pakistan and is cultivated on large area. Mealy bugs, hoppers, scales and midges are the insect pests of mango but fruit fly (Bactocera zonata) is an important insect pest of mango fruit and belongs to the order Diptera and family tephritidae. Now a days it is major barrier in successful production and causes serious mango losses due to its wide distribution, fast propagation and polyphagous nature. The present experiment was conducted to evaluate the efficiency of pheromonal and electric traps against B. zonata in mango orchards at District Khanewal. Two types of traps pheromonal and electric traps were used in an acre. Two concentrations 4ml and 2ml of methyl eugenol were used at two heights of 5 feet and 3 feet in pheromonal traps and data were collected on daily basis. Also, electric light trap was applied at two different heights of 5 feet and 3 feet. Data were collected on daily basis for 20 days. Average 32 fruit flies per traps of 5 feet height were recorded on 4ml concentration of methyl eugenol and average 24 fruit flies were collected per trap of 3 feet height on 2ml conc. of methyl eugenol. There was no effect of electric traps against fruit fly but it was affected against hoppers and other pests of mango. Results indicate that pheromonal traps with more concentration of methyl eugenol and with more height were very effective against fruit flies in mangoes. For the more production of mango yield farmers should use the more concentration in traps for the management of fruit fly.

# POPULATION PHENOLOGY OF PINK BOLLWORM (*PECTINOPHORA GOSSYPIELLA*) ON TRANSGENIC AND NON-TRANSGENIC COTTON UNDER SPRAYED AND UNSPRAYED CONDITION

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The pink bollworm is the most important pest of cotton specie *gooypium* in the cotton cultivation zones all over

the globe. The control of *P. gossypiella* on cotton has become a tough task because it has developed resistance against transgenic cotton. The *P. gossypiella* spend their developmental stages within the cotton bolls therefore, conventional control methods including insecticidal applications are not very effective to control the pest. The cultural control plays a primary role in decreasing the carry-over of p. gossypiella population between cotton crops. Therefore, the study was conducted to understand the seasonal incidence of P. gossypiella on transgenic and non-transgenic cotton under the sprayed and un sprayed conditions, for better understanding of the pest. The number of *P*.

gossypeilla was recorded from the green bolls with the interval of one week from September to October. In total, 25 plants were selected on random basis from each sprayed and unsprayed transgenic and non- transgenic plots to record pest population. Date regarding number of infested bolls, number of P. gossypiella larvae/ bolls number of single seeded diapausing larvae and double seeded diapausing larvae and; larval instars during cropping season and off season was recorded and percentage infestation counted. The presence of p. gossypiella was detected in both sprayed and unsprayed transgenic and non- transgenic cotton field. The number of infested bolls was lowest on the sprayed transgenic cotton 4.25 bolls /25 and highest on unsprayed non- transgenic cotton 13.78 bolls/25 bolls. The number of P. gossypiella was lowest on sprayed transgenic cotton 1.81 larvae/ 25 bolls and highest on unsprayed non-transgenic cotton 8.14 larvae /25 bolls. The number of double seeded diapausing larvae was lowest on sprayed transgenic cotton 1.92 larvae/25 bolls. The number of single seeded diapausing larvae was lowest on sprayed transgenic cotton 1.45 larvae/25 bolls and highest on unsprayed non-transgenic cotton 6.42 larvae/25 bolls. The infestation of P. gossypiella during off season was lowest on sprayed transgenic cotton 6.71% and highest on unsprayed non-transgenic cotton 34.67%. This can help to formulate control strategies and evaluate new insecticide molecules and techynologies for effective management of P. gossypiella.

## MANAGEMENT OF PINK BOLLWORM (*PECTINOPHORAGOSSYPIELLA*) IN COTTON GINNING WASTE BY ENTOMOPATHOGENIC FUNGI

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Ginning waste contains organic matter which can be amended in the soil for the upsurge of soil productivity. It contains nitrogen which is primary essential for plant growth, small amount of potassium and phosphorus. Same while ginning waste is also called as nursery of pink bollworm (PBW), a major pest of the cotton. The present study was conducted to control the PBW in ginning waste by using entomopathogenic fungi *Beauveria bassiana* and *Materhzium anisophilae*. The 20g of ginning waste and 10 PBW third instar larvae per petri dish was used in each treatment. In liquid formulation distilled water was used as a carrier and for the dry formulation autoclaved clay was used. Seven treatments were used with five replications. Two concentration of *B. bassiana* (1mg\10ml,1.5mg\10ml),two of *M.anisophilae*(1mg\10ml, 1.5mg\10ml), two of dry formulation of *B. bassiana* and *M.anisophilae*was applied @1.0mg\10mgand last was untreated control. Larval, pupal and adult mortality was observed after 4<sup>th</sup>, 7<sup>th</sup>, 14<sup>th</sup> and 21<sup>st</sup> day of treatment. Maximum larval mortality was observed at *B. bassiana* liquid concentration 1.5mg\10ml, while maximum pupal morality was observed for *M. anisophilae* at 1.5mg\10ml concentration. It was observed that EPF are effective management strategy for PBW in ginning waste.

# SUB-LETHAL EFFECTS OF FLUBENDIAMIDE (BELT $^{TM}$ ) ON ADULT LONGEVITY OF SPODOPTERA LITURA IN LABORATORY

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Spodoptera litura Fab., (Lepidoptera: Noctuidae) is one of the most dangerous insect pests which attack on 40 plant families and about 150 species of different plants. Present, studies were conducted to evaluate the sub-lethal effects of flubendiamide (Belt<sup>TM</sup>) on adult longevity of *S.litura* in laboratory conditions at Department of Entomology, Sindh Agriculture University, Tandojam at 26 °C and 60% R.H. The larvae were collected from vegetable fields of Tando Allahyar and reared on cauliflower leaf discs. Cauliflower leaf discs were dipped in five different water concentrations of insecticide as 57.6, 48, 38.4, 28.8 and 19.2 ppm. Treated leaf discs were provided to fourth instar larvae. Surviving larvae after 48 h were provided untreated leaf discs and sub-lethal effects of

flubendiamide were assessed on adult longevity. Results indicated that maximum longevity  $7.00 \pm 2.88$  was observed at 57.6 ppm followed by  $6.96 \pm 0.20$  at 48 ppm,  $6.60 \pm 0.45$  at 38.4 ppm,  $6.6 \pm 0.4$  at 28.8 ppm,  $6.6 \pm 0$  at 19.2 ppm and  $10.1 \pm 0.5$  at control. It is concluded that adult longevity of *S.litura* was decreased on maximum doses and increased at lower doses as compared to control.

### TRITROPHIC INTERACTION AMONG ALTERNATE HOSTS COTTON MEALYNUG, PHENACOCCUS SOLENOPSIS (PSEUDOCOCCIDAE: HEMIPTERA) INVOLVING COCCINELLA SEPTEMPUNCTATA (COCCINILLIDAE: COLEOPTERAN)

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Because of their differential morphological and biochemical traits, host plant genotypes have pronounced effects on the biology of pests and related entomophagous insects through tritrophic interaction. Understanding interactions among host plants, herbivore and the predators are important for the development of optimal integrated crop management strategies, but empirical studies investigating such tritrophic interactions were uncommon. In the present studies, the effects of some host plants on Coccinella septempunctata and cotton mealybug were evaluated. The experiment was conducted in IPM (Integrated Pest Management) laboratory, Department of Agri, Entomology, University of Agriculture, Faisalabad. It was recorded that: maximum percentage mortality was observed in third instar of mealybug on Rose (88.89%) and minimum mortality in third instar of mealybug was observed on shoeflower (11.11%). Nymphal duration of cotton mealybug was prolonged when fed on mango (23.33 days) while minimum nymphal duration was observed on Shoeflower (424.245 crawler/ovisac and minimum crawlers per ovisac of cotton mealybug was produced by cotton mealybug female when it was fed on rose (150 crawler/ovisac).Maximum mortality was observed in C. septempunctata when fed on Jatropha (86.66%) and minimum percentage mortality in C. septempunctata when fed on Cotton (87.33%) and minimum percentage food consumption was observed when fed on Niazboo (34%).

# EVALUATION OF INDIGENOUS MEDICINAL PLANTS AS TOXICANTS AND REPELLENTS AGAINST INSECT PEST OF STORED GRAINS, RHYZOPERTHA DOMINICA

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The objective of the study was to appraise the insecticidal bioactivity of medicinal plants; *Moringa oleifera* (Sohanjana), *Datura stromonium* and *Citrullus colocynthis* (Tuma) against most destructive primary insect pest of stored grains i.e. lesser grain borrer, *Rhyzopertha dominica* Everts (Coleoptera: Bostrichidae). Bioassays were carried out to examine the toxicity of plant extracts at four dose rates *viz*; 2.5%, 5.0%, 7.5% and 10.0% for mortality and repellency against *R. dominica*. The outcomes evidenced that mortality of 5 days old larvae of *R. dominica* increased with dilution level from 2.50 % to 10.0% and with time exposure from 2 to 10 days. Plant extract of *C. colocynthis* at 10.0% concentration reported maximum mortality (46.06%) after 10 days interval, while other two plants *M. olifera* and *D. stromonium* forced a maximum mortality of 37.07% and 30.33%, respectively at 10.0% dose rate after 10 days. For deterrence effect, filter paper treated with 10.0% extract of *C. colocynthis* evidenced maximum repellency (91.11%) from three plants after 3 days interval. These findings suggested that naturally occurring plant extracts have decent insecticidal potential for the control of *R. dominica* and can be used in the IPM program of stored grains pest management.

### EFFECTS OF INTEGRATION OF NUTRIENTS AND INSECTICIDES ON SUCKING INSECT PESTS OF COTTON IN FAISALABAD

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Several insect pests attack on cotton crop. Due to injudicious use of pesticides; the human health as well as our environment have been severely deteriorated. In order to reduce the usage of pesticides to control insect pests, economical and environmentally friendly techniques should be applied. The attack of insect pests increases in nutrient deficient soils. The present study was conducted to evaluate the combined effects of nutrients and insecticides in population build-up of sucking pests in cotton. Research was conducted under Randomized Complete Block Design. Three treatments each with three replications were applied. The data of sucking insect pests was collected on daily basis and analyzed by ANOVA technique. The results showed that nutrient formulations of Ali Akbar group integrated with insecticides showed best results against sucking insect pest population and increased the yield. The impact of nutrients of Warble group was also observed and compared with Ali Akbar group.

### POULTRY: A NATURAL TOOL TO CONTROL THE TICKS IN LIVESTOCK FARMS

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Ticks are one of the major problems and threat to the livestock industry globally. They are obligate ectoparasites and feed on the host blood. They are considered as the second biggest vectors of human disease worldwide. In European countries, more the 50,000 cases of tick-borne disease (TBDs) are reported annually. They influence the animal health directly or indirectly. In livestock, their biting cause inflammation, hypersensitivity, irritation, weakness, 1-2 kg (or 20 kg in a year) weight loss, anemia or even death of the host in severe cases. High infestation is the reason of reduction in milk, meat and other livestock origin products. A single female can cause 8.9 ml deduction in milk production. According the research report, globally 80% and 50% losses occur in cattle and sheep population respectively. Wounds due to their biting cause attraction of other pathogens and parasites. Ticks are easily transmitted from infested animal to healthy one by contact or movement. The same and incorrect use of chemicals for the control of ticks is the basic reason of resistant and tolerance in ticks which make difficult to control them on recommended dose. Among the biological control poultry, mostly hens, are found a natural source of ticks control. They eat a larger number of ticks with the average ingestion rate 28-81 ticks per chicken reported in Kenya and South Africa respectively. Locally, we also found that, the farmers who reared poultry on their livestock farm have less infestation of ticks on their animals. Therefore, it is concluded that, rearing of chickens is the easiest and cheapest tool to control the ticks and TBDs naturally.

### CURRENT SCENARIO OF WHITEFLY, BEMISIA TABACI GENN. IN DIFFERENT COTTON GROWING AREAS OF SINDH-PAKISTAN

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Cotton is one of the most main industrial crops in Pakistan and worldwide. Cotton is a major fibre crop of global importance and has high commercial value. It is grown commercially in the temperate and tropical regions of more than 70 countries. Specific areas of production include countries such as China, USA, India, Pakistan,

Uzbekistan, Turkey, Australia, Greece, Brazil, Egypt etc where climatic conditions suit the natural growth requirements of cotton. Sindh province enjoys warm sunny climate and is well suited to cotton cultivation. An entomological survey of five years on pest scouting was conducted to evaluate the current scenario of whitefly pest position on cotton crop in Sindh Province of Pakistan. The data shows that in last five years whitefly population on cotton is increasing day by day in Sindh. The data in the current scenario indicate that whitefly has become a big threat on cotton and giving the qualitative and Quantitative loss to the cotton crop in the Sindh Province. During the survey growers told that we applied the different groups of pesticides of different companies for the control of these pests but applied pesticides did not given the satisfactory results for the control of whitefly. Further results shows that growers mainly depends on insecticides and due to the indiscriminate use of insecticides it also create many other problems like, destruction of natural enemies of the pest, resurgence of secondary pests, development of resistance. A apart from this indiscriminate use of insecticides also create environmental pollution too. The results showed that maximum whitefly population was recorded in Ghotki, Sukhur and Khaipur and Nausharo Feroze districts, respectively. While minimum population was observed in Badin, Tando Muhammad Khan and other districts of Sindh Province. Keeping in view the above mentioned issues of whitefly and its presence throughout the year, it needs care full attention. It is therefore suggested that on the basis of results Govt. and Research Organizations should made the research policy to control this pest in proper way to save the cotton crop as well as economy of the Country.

# EFFICACY OF DIFFERENT INSECTICIDES FOR THE CONTROL OF MANGO THRIPS SCIRTOTHRIPS DORSALIS HOOD. (THRIPIDAE: THYSANOPTERA) AT NURSERY LEVEL

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Mango thrips Scolothrips dorsalis Hood, is known for one of the major pest in upcoming future because it is not only sucking the cell sap of the leaves, but lacerated the leaf and fruit surface as well as deteriorates the mango fruits at stone stage and above. While in severe attack small stone size fruits dropped and large fruits are affected with large blemishes showing corky dark rusty stains. So to avoid such kind of damages at nursery level, a study was conducted to evaluate the efficacy of different insecticides commercially available in market for the control of this notorious pest. Equal sized nursery blocks with young nursery plants were initially selected which were infested with thrips. Nursery plants were divided into 14 different blocks in order spray. 13 different insecticides were selected and applied according to the recommended doses given by the label at nursery level. Manually operated knapsack sprayer was used in order to spray insecticides. Data was recorded randomly before and after 24h and 72h of spraying insecticides. Simple hand lens was used to count the thrips underside the fresh leaf blushes. The results showed that percent mortality of thrips as descending order from maximum to minimum; Chlorfenapyre< Imidacloprid + Acetamaprid < Imidacloprid < Nitenpyram+Chlorfenapyre<Diafenthiron<Thiamethoxam<Triazophos<Bifenthrin< Nitenpyram+Pymetrozine<Fipronil<Clothioindin<Malathion<Sulfoxaflor. Percent mortality of thrips population after 24h and 72h of spraying insecticides was recorded as 56.72<55.65<42.27<32.65<29.75<25.44 <18.49<16.70<9.85<5.60<5.35<3.97<2.49 and 53.64<47.4 <37.63<19.23<13.8<16.53<14.67<14.26<4.71<4.49 <8.75<2.67<3.65 respectively.

# ENTOMOPATHOGENIC NEMATODES STEINERNEMA CARPOCAPSAE AND HETERORHABDITIS BACTERIOPHORA AFFECT FITNESS OF A VIP3A RESISTANT SUB-POPULATION OF HELIOTHIS VIRESCENS

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Resistance management to insecticides is widely believed to depend in part on associated fitness cost. In the present study the effects of two entomopathogenic nematode species, *Steinernema carpocapsae* and *Heterorhabditis bacteriophora* were studied on the fitness of first insect population of *Heliothis virescens* selected with Vip3A in the laboratory. The result indicated that both the nematodes species increased the fitness cost of Vip3A selected insects. The mortality of the Unsel sub-population after exposure to either nematode species was significantly lower than the Vip3A-Sel sub-population. Likewise, the reproduction of both the nematode species was significantly greater in cadavers of the Unsel compared with the Vip3A-Sel sub-population of *H. virescens*. There was positive correlation between nematode reproduction and the larval instar infected with nematodes. The penetration of IJ was greater in the Vip3A-Sel sub-population than in the Unsel sub-population of *H. virescens*. It is concluded that entomophatogenic nematodes could increase the fitness costs and subsequently delay the resistance.

# VULNERABILITY OF COTTON MEALY BUG, PHENACOCCUS SOLENOPSIS (TINSLEY) TO DIFFERENT ENTOMOPATHOGENIC FUNGAL STRAINS AT DIFFERENT DEVELOPMENTAL STAGES

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Present research work was carried out to evaluate the pathogenic potential of entomopathogenic fungi (B. bassiana, M. anisopliae and V. lecanii) against different development stages of P. solenopsis under laboratory conditions. Fungus strains were purchased from Agri-life India and evaluated against different life stages of CMB. Results revealed that B. bassiana caused 50% mortality ( $LC_{50} = 7.89 \times 10^5$  spores/ml,  $LC_{50} = 1.24 \times 10^6$  and  $LC_{50} = 1.24 \times 10^6$  $4.87 \times 10^6$ ) for 1st instar, 2nd instar and adult female of cotton mealybug respectively. Similarly M. anisopliae exhibited (LC<sub>50</sub> =  $5.54 \times 10^6$  spores/ml, LC<sub>50</sub> =  $1.98 \times 10^6$  and LC<sub>50</sub> =  $8.41 \times 10^7$ ) for 1<sup>st</sup> instar, 2<sup>nd</sup> instar and adult female of CMB respectively. V. lecanii tested against P. solenopsis, showed LC<sub>50</sub> value of  $5.85 \times 10^6$ ,  $3.2 \times 10^6$  and  $8.87 \times 10^7$  on first instar, second instar and adults respectively. The LT<sub>50</sub> ranges for different concentrations of B. bassiana were 3.23 to 4.23 days against 1st instar, 3.81 to 4.63 days against 2nd instar and 4.32 to 5.56 days against adults in the P. solenopsis. While LT<sub>50</sub> ranges for different concentrations of M. anisopliae were 3.88 to 4.67 days against 1st instar, 3.98 to 4.89 days against 2nd instar and 4.88 to 5.77 days against adults of CMB. However LT50 ranges for different concentrations of V. lecanii were 4.54 to 4.98 days against 1st instar, 4.45 to 5.62 days against 2nd instar and 5.55 to 6.88 days against adults of CMB. In the present study, the LC<sub>50</sub> and LT<sub>50</sub> values indicated that the reduction in virulence with the advancement of the developmental stages. The susceptibility of different stages of the mealy bug to the pathogen and its ability to transmit infection among the various developmental stages, lend support to the potential of entomopathogens for biological control of mealy bug.

### IMPACT OF DIFFERENT ADULT-DIETS ON THE SURVIVAL AND REPRODUCTIVE POTENTIAL OF BACTROCERA ZONATA

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Bactrocera zonata is highly injurious pest of fruits, vegetables and is responsible for huge economic losses in term of qualitative and quantitative damage all over the world. present study was carried out to evaluate the impact of different adult diets on the survival and reproductive potential of B. zonata. The study was consisting of two experiments. In the first experiment, eight types of fruit fly diets were prepared and offered to pre-determined number of pairs of B. zonata in separate feeding units by surface pasted diet method. Honey solution was used as a stander adult diet for comparison and water-soaked sponges were used as a control treatment. The emerged adult flies aspirated from culture manually, kept starved for two hours and then released in feeding units. The feeding units were also provided with oviposition substrate/receptacle fruit for collection of eggs. After a period of two days exposure, the results demonstrated that maximum number of larvae (32.23larvae) and pupa (29.30 pupa) was recorded on diet-H as compare to other diets to determined oviposition. The mortality data were counted on daily basis till all flies died. The maximum longevity of female (30,00 days longevity) was recorded on diet-A Oviposition period (14.91 days) and pre-oviposition period (25.02 days) fecundity/female, fecundity/female/day, mortality rates per day (5.00) was also recorded or computed. The impact of these diets was significant. In second experiment, the highly effected diet (diet-H) was admixed with juices of ten fruits and results demonstrated that maximum longevity (41.00 days) was observed on diet-D and diet-G respectively.

### INTEGRATED MANAGEMENT OF TROGODERMA GRANARIUM USING NEEM LEAF EXTRACT, DIATOMACEOUS EARTH AND ENTOMOPATHOGENIC FUNGUS

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Infestation by different insect pest to stored cereals and their products storage is a serious problem. Khapra beetle (Trogoderma granarium) is highly destructive insect pests of stored cereal products, causes both the nutritional and economical losses to stored cereals. This present study work were conducted to evaluate the integrated effect of different protectants like plant extract (neem), diatomaceous earth (DE) and entomopathogenic fungus (Beauveria bassiana) against Trogoderma granarium. Six different concentrations of each protectants were used for the bioassay. For neem leaf extract dose rate were (5%, 10%, 15%, 20%, 25% & 30%) leaves were collected from University of Agriculture Faisalabad, Pakistan, for DE formulation (Conern®) doses rates (200ppm, 400ppm, 600ppm, 800ppm, 1000ppm & 1200 ppm/kg of wheat grains) and (1g, 2g, 3g, 4g, 5g & 6g/kg of wheat grains) doses of Beauveria bassiana (Racer Bb) (1 g of Racer containing 108 conidia) was imported from Agri Life, Medak District. Hyderabad, India were used. 30 larvae will used in each bioassay. There were three replications of each treatment. While data for Diatomaceous earth and neem leaf extract and entomopathogenic fungus were recorded after 7, 14 and 21 days. All data were computed statistically using statistica-10 software and corrected mortality of recorded data was calculated using Abbott's formula (Abbot, 1925). Mean % mortality of test insect increases by increasing the concentration and exposure time (days). Maximum mortality (62.20%) was observed after 21 days, After 14 days (42.29%) and after 7 days (24.05%). Minimum mortality was found after 7 days. The present study revealed that the information about the combination of neem plant extract, Diatomaceous earth and entomopathogenic fungi (Beauveria bassiana) can serve as guideline for adopting effective measure against T. granarium.

### CONTROL OF MOSQUITO-BORNE DISEASES: *VIA* THE *WOLBACHIA*-BASED INTERVENTIONS IN AN INTEGRATED VECTOR MANAGEMENT FRAMEWORK

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Mosquitoes pose a major health issue to the millions of people, either living in tropical or sub-tropical regions of the world. Just because of the disease spread by the mosquitoes, one million people die each year. The notable diseases are spread by it such as malaria, dengue fever and filariasis which have a direct impact on the human health. As of today, and the deep look of past, the only method in use to control the mosquitoes are the use of insecticides (chemicals). Though, use of the mentioned technique has created many questions in terms of resistance to mosquitoes. Thus, moving towards the innovative and promising tools is the need of time. So, in this review a comprehensive background of the new emerging technologies is presented such as the method of Trans-infecting. In this one, the vectorial capacity of mosquitoes will be slow down by using the targeted pathogens *i.e. Wolbachia*. Furthermore, other tactics to control the mosquitoes-borne diseases by using mosquitoes is presented.

### ASSESSMENT OF ENTOMOPATHOGENIC FUNGI BEAUVERIA BASSIANA WITH INTEGRATION OF BACILLUS THUREINGENSIS AGAINST JASSID POPULATION IN OKRA

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Okra is an important vegetable crop grown in Pakistan. Quality of fruit and its production was reduced by attack of several insect pest. Pesticides were used to control these insect pests around the globe. Pesticides have adverse effect on the human health as well as toxic to environment. So, the present experiment was designed to evaluate the potential impact of *Beauveria bassiana* and *Bacillus thureingensis* against population of Jassid in okra in field condition. All treatment was designed in randomized complete block designe (RCBD) with three replications of each. Population reduction data were collected after treatment application. Result showed that Jassid population reduction was maximum recorded in case of *B. bassiana* treated blocks about 78% while minimum population were observed in *B. thureingensis* treated blocks. Combined application of *B. thureingensis* and *B. bassiana* showed maximum reduction in population about 85% whereas minimum reduction was found in case of *B. thureingensis*. It can be concluded that entomopathogenic fungi and *B. thureingensis* could be effective to control Jassid population in okra and which are ecofriendly to environment.

### ASSESSMENT OF ATTRACT-AND-KILL POTENTIALS OF DIFFERENT FOOD BAITS AGAINST TEPHRITID FRUIT FLIES

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Tephritid fruit flies are one of the serious threat to the horticultural crops and causes severe losses to economics. Present study was carried out for the assessment of attract-and-kill potentials of different food baits against *Bactrocera zonata*, *B. dorsalis* and *B. cucurbitae*. Different food bait formulations were prepared and applied in

plastic bottle traps for their respective response. Traps were installed in guava orchard Square-9 and bitter gourd field at Entomological area, University of Agriculture, Faisalabad. Data were collected at two days intervals for one month for each experiment. All experiments were followed by RCBD with three equal replications. The collected data were subjected to ANOVA. Tuckey HSD test was used in case of significant results to compare the means at 5% probability level. Food-lures (FLs) were prepared by admixing food supplements containing banana, protein hydrolysate, mulberry syrup, molasses, guava, coconut-oil and grapes extract with different combinations. However, GF-120, banana paste, and protein hydrolysate were used as standards for comparison (experiment-1). Results revealed that FL-6 proved 33.54%, 26.73% and 24.90% (Class-II); 31.55%, 33.57% and 15.89% (Class-II); 12.57%, 49.58% and 36.10% (Class-II) more attractive to male B. zonata, B. dorsalis and B. cucurbitae over standard-1, standard-2 and standard-3 respectively while FL-6 proved 33.69%, 29.74% and 27.90% (Class-II); 24.82%, 20.73% and 25.77% (Class-II); 10.46%, 39.00% and 28.88% (Class-II) more attractive to female B. zonata, B. dorsalis and B. cucurbitae over standard-1, standard-2 and standard-3 respectively. Based on the 1st experiment results, highly attractive food lure (FL-6) was selected and admixed with different combinations of ammonium acetate, trimethylamine, putrescene, rose oil and DAP for food-lure-mixtures (FLMs). However, protein hydrolysate and GF-120 were used as standards for comparison (Experiment-2). Results revealed that FLM-12 proved 66.64% and 55.24% (Class-III); 51.94% and 56.87% (Class-III) more attractive to male and female B. zonata over standard-1 and standard-2 respectively, FLM-3 proved 55.42% (Class-III) and 33.04% (Class-II); 46.16% and 47.68% (Class-III) more attractive to male and female B. dorsalis over standard-1 and standard-2, FLM-12 and FLM-18 proved 57.78% and 56.63% (Class-III); 52.54% and 51.29% (Class-III) more attractive to male and female of B. cucurbitae over standard-1 and standard-2 respectively. Multi-lure mixtures (MLMs) were prepared by mixing three highly attractive FLMs (FLM-3, FLM-12 and FLM-18) from 2<sup>nd</sup> experiment. These MLMs then were admixed with equal concentration of insecticide (Spinosad) in each trap to check the Attract-And-Kill potentials against fruit flies (Experiment-3). Results revealed that MLM-3 proved 43.33%, 41.15% and 41.40% (Class-II); 59.60%, 68.28% and 57.59% (Class-III) more attractive to male and female B. zonata, B. dorsalis and B. cucurbitae respectively than other lures. MLM-3 proved highly attractive multi-food-lure-mixture and showed greater attract-and-kill potentials against fruit flies. Use of different food bait combinations proved effective and environmentally safe control of different fruit fly species.

### FRUIT FLY A MAJOR MANGO FRUIT THREATEN AND THEIR FUTURE PROSPECTS RELATING WITH ENVIRONMENTAL CHANGE IN TROPICS AND SUB-TROPICS

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Mango (Mangifera indica L) is economically significant fruit and shared a large amount in gross domestic product (GDP) by exporting every year. In Pakistan amongst the major threat to mango, fruit fly (Diptera: Tephtritidae) is considered as alarming pest by causing a huge loss. This pest is cosmopolitan especially in tropical and non-tropical areas of the world. Mainly two mango fruit fly families including Bactrocera Zonata and Bactrocera dorsalis found in Pakistan. From the last decades it caused a major loss nearly 30% in the mango orchard in Pakistan. Maximum attack in mango orchard were seen when fruits reach at the semi ripen and mature stage. Environmental conditions also play a major role in pest infestation rate. According to present climatic situations, its mainly potential threat was in tropics and sub-tropics. But depending upon climate change it is predictable that this pest distribution become widespread. Though, it is necessary no know the potential threat of this pest and make a strategy according to climate change. So, in Pakistan this pest will become more potential by depending upon the environmental conditions. The impact of this pest is striking and habitat range will be increased potentially specifically in Asian countries. Therefore, it is necessary to spread proper knowledge about climate change relating with fruit fly potential and use the IPM technique for control.

### SYNERGISTIC TOXICITY OF PESTICIDE MIXTURES TO CONTROL TROGODERMA GRANARIUM: EFFECT ON ENERGY RESERVES

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Trogoderma granarium is an important quarantine insect pest of amylaceous products. Its control has become difficult due to the emergence of resistance against almost all the known insecticides and fumigants. The development of resistance can be delayed by using binary combinations of insecticides. Keeping in view the importance of synergism as well as the concerns about environmental hazards and emergence of resistance against these insecticides, the current research was designed to figure out the lethal concentration (LC50) of emamectin and deltamethrin alone and in various combinations such as emamectin:deltamethrin (1:1, 2:1, 3:1, 1:2 and 1:3) against two larval instars (4<sup>th</sup> and 6<sup>th</sup>) of Lahore population of T. granarium. The LC<sub>50</sub> of emamectin and deltamethrin for 4<sup>th</sup> larval instars were 57 and 81 ppm, respectively while LC<sub>50</sub> of these insecticides against 6<sup>th</sup> larval instar of was 51 and 78 ppm, respectively. The emamectin was the most effective than deltamethrin. Based on relative toxic unit, the 3:1 mixture of emamectin:deltamethrin showed higher toxicity among all the tested mixtures. The toxic effect of LC<sub>20</sub> of this 3:1 mixture on soluble and total proteins, free amino acid, total lipids, glucose, glycogen and trehalose contents was also recorded. The results indicated that contents of glycogen, trehalose, total lipids, free amino acids, soluble proteins and total proteins were significantly deceased in both larval instars. The glucose contents increased in both larval instars of exposed groups with reference to unexposed groups. It is concluded that the mixtures of insecticides were more effective than the administration of insecticides alone to control T. granarium.

### PESTICIDES BIOREMEDIATION USING MICROORGANISMS; AN ECO-FRIENDLY APPROACH

#### Manam Walait and Mushtaq A. Saleem

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The use of pesticides in agricultural fields is a major concern now-a-days as pesticides are involved in causing serious issues to human health and environment. Bioremediation of the pesticides is a promising environment friendly technology that has gained attention of researchers across the globe from the past few decades. Bioremediation utilizes naturally occurring or deliberately introduced microorganisms to reduce, remove, degrade or immobilize environmental toxins, in order to clean polluted site so that polluted sites can be stored to a condition useable for private or public applications. Some of the pesticides such as insecticides, rodenticides, herbicides and chlorinated compounds are highly toxic and lethal even in trace amounts. In Pakistan, endosulfan and atrazine has been used widely as insecticides and herbicides respectively for many years. Persistence of these pesticides in soil helps in their biodegradation. Bioremediation has been found effective in remedying the high concentration of these pollutants from the soil and ground water. Some microorganisms have interestingly been found effective in absorbing toxic elements from air as well. Soil microorganisms have ability to degrade atrazine to a non-toxic metabolite. Some of the bacterial strains such as Arthrobacter sp., Chelatobacter heintzii, Rhodococcus sp., Acinetobacter sp., Streptomyces sp., Pseudomonas aeruginosa, Clavibacter michiganens, Enterobacter cloacae, Bacillus megaterium, Alcaligenes faecalis, Klebsiella ornithinolytica, and Agrobacterium tumefacien were reported as efficient strains which can degrade pesticides into its metabolites. Besides bacterial strains, many fungal strains e.g. Aspergillus sp., Trichoderma sp., Fusarium sp., Rhizopus sp., and Penicillium sp. were also found important for degradation of pesticides The lab-scale strategies should be applied on field so that outcomes of these strategies should be observed and more improvements in these procedures should be made for better acquisition of these bioremediation technologies.

#### POTENTIAL CONTROL METHODS OF FRUIT FLIES (DIPTERA: TEPHRITIDAE) IN PAKISTAN

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Eleven species of fruit flies (Diptera: Tephritidae) are reported (e.g., from Azad Jammu and Kashmir, Gilgit Baltistan, Sindh, Punjab, Khyber Pakhtunkhwa and Baluchistan provinces) as pests of fruits and vegetables in Pakistan where *Bactrocera zonata*, *B. dorsalis* and *B. cucurbitae* considered to be most destructive but *B. dorsalis* is evaluated as a major quarantine pest threat in foreign export of fruits and vegetables especially thorough China Pakistan Economic Corridor (CPEC). This review highlights the importance of control practices either pre- and/ or post harvest that were combined after reviewing more than 100 fruit flies related articles. We observed following management practices being experimented or practically applied in the form of field sanitation by burying fruits three feet deep inside the soil, bait application technique (BAT) e.g., steiner traps baited with methyl eugenol (attractant), parasitoids and predators (ants, ground beetle and spiders), botanicals, insecticides and bio rational insecticides (e.g., spinosad) by different colour refractive sheets. One of the main factor lacking in Integrated pest management is the growing of less susceptible varieties. Post-harvest management include hot water treatment, cold treatment, irradiation (X-rays, gamma ray, etc.), vapour heat treatment, and male annihilation technique (MAT). Overall, an area-wide integrated pest management (AW-IPM) strategy is being experimented to aware the farming community. In conclusion, given above information would provide opportunities to those who are working on the sustainable management of fruit flies that is need of time to implement in Pakistan.

#### MANAGEMENT OF WHEAT APHIDS BY USING CAMELINA SATIVA AS TRAP CROP

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For better wheat productivity, wheat aphids cannot be controlled by using insecticides as wheat is stable food in Pakistan. It could be managed by using push and pull strategy through enhancing natural enemy population. In the current study, camelina was planted as trap crop with objectives to check its potential as trap crop. The total experimental area was 0.036 ha (wheat alone, camelina alone and wheat sand-witched between camelina plots) at Entomological Research Area of University of Agriculture Faisalabad, Pakistan during one season study in 2018-2019. Each plot was sub divided into four sites and data was taken by five fixed sampling method in open field survey through visual observations. Experiment was conducted at seven days' interval from January 29th to April 23th, 2019. There was no use of pesticides and weeds were removed manually. As a result, highest mean number (66.44) natural enemies (coccinellids, syrphids, araneae and parasitoids) and highest mean number of aphids (320.70) per 100 tillers were observed in alone wheat plots when compared with wheat sand-witched between camelina plots where significant lowest mean number of aphids (100.59) and lowest mean number of natural enemies (66.07) per 100 tillers were reported. In case of camelwina, highest mean number (99.06) natural enemies (coccinellids, syrphids, araneae and parasitoids) and aphids (280.39) per 100 plants were observed in wheat sand-witched between camelina plots when compared with camlina alone plots where lowest mean number of aphids (207.37) and natural enemies (66.26) per 100 plants. Overall, finding manifested that camelina have precise qualities of trap crop which act as a best reservoir for natural enemies and the diversity of predators and parasitoids facilitates the non-chemical control of wheat aphids. These finding would be very helpful for farmer community to take pre planned management actions before pest outbreak.

### IMPACT OF BOTANICAL EXTRACT AGAINST JASSID, AMRASCA BIGUTTULA BIGUTTULA (ISHIDA). ON SUNFLOWER CROP

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The experiment was conducted during 2018 at the field of Entomology section Agriculture Research Institute Tandojam on the impact of botanical extract against jassid, *Amrasca biguttula biguttula* (Ishida). on sunflower crop using of bio-pesticide neem (*Azadirachta indica*), datura (*Datura stramonium*) and eucalyptus (*Eucalyptus terticornis*) while their impact was compared with an untreated control plot was also maintained. Results indicated that overall maximum mean population (2.95±0.19) was recorded in datura followed by (2.68±0.20) in eucalyptus and neem (2.47±0.15). The reduction percent of jassid indicate that overall maximum mean reduction (51.39±2.93) was recorded in neem followed by (42.80±5.32%) in datura and (33.44±4.98%) in eucalyptus. The ANOVA results showed that treatment means varied significantly at (P= 0.05) level. LSD confirmed that two separate groups A, B and C were found showing better performance of datura bio-pesticides than other bio-pesticides. The maximum yield obtained from T2 (datura) followed by T1 (neem) and T3 (eucalyptus) compared with T4 (control plot).

## BIOLOGICAL EFFECTS OF PLANT POWDERS AGAINST MAIZE WEEVIL (SITOPHILUS ZEAMAIS), MOSTCH (COLEOPTERA: CURCULIONIDAE)

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Biological screening of six plant products including Neem seed (*Azadirachta indica*), Bitter cress succulent fruit (*Caralluma turberculata*), Garlic rhizomes (*Allium sativum*), Turmeric rhizome (*Curcuma longa*), Tumha fruit (*Citrullus colocynthis*) and Ak leaves (*Calotropis procera*) was carried out in powder form against maize weevil, a cosmopolitan pest of stored maize. The studies were carried out following completely randomized design having 5 repeats in the laboratory of Entomology Department, Gomal University, Dera Ismail Khan, KP Province, Pakistan to investigate the effect of selected plant powders on the days to first filial generation (F1) emergence, total number of adults emerged, percent infestation and weight loss, sex ratio and longevity of F1 adults. Results indicated that all the evaluated plant powders had significant (P<0.05) effect on the biology of maize weevil compared to control. Among the tested plant powders, *A. indica* and *C. longa* were found more effective for the management of maize weevil as they showed minimum infestation (8.69 and 13.87%,) weight loss (3.63 and 5.86%, less adult emergence (21.43 and 36.60) and extended the duration of F1 emergence (38.33 and 36.70 days) of the test insect. Henceforth, it is concluded that the selected plant materials could be used effectively for the management of maize weevil.

### ACARICIDAL POTENTIAL OF *EUPHORBIA HELIOSCOPIA*, *EUPHORBIA HIRTA* AND *BUXUS WALLICHIANA* ALCOHOLIC EXRACTS AGAINST *HYALOMMA* TICKS

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Ticks and tick-borne diseases (TTBDs) affect 80% of the world cattle population and are widely distributed throughout the world. In Pakistan piroplasmosis, theileriosis and anaplasmosis are common livestock diseases, which are transmitted by the ticks, which results in increased irritation, restlessness, weight loss and disease transmission. The present study was aimed to evaluate toxic activity of *Euphorbia helioscopia*, *Euphorbia hirta and Buxus wallichiana* leaves extract against genus *Hyalomma* ticks. The leaves of these three plants were collected from different areas of Khyber Pukhtunkhwa, Pakistan. The leaves of these selected plants were dried for few days and then leaves were crushed in electric grinder. The selected plant powder were shaked in ethanol solvent. After that filtered extract of selected plants were prepared. For determination of toxic activity of these three plants, *Hyalomma* ticks were placed in each petri dish having different concentrations (10, 30, 50 mg/ml) of ethanol extracts of leaves. Percent mortality of ticks was calculated after 2 hours. Toxicity effect of the 3 extracts against *hyalomma* ticks detected was in subsequent order, *Euphorbia helioscopia* > *Euphorbia hirta* > *Buxus Wallichiana*. This may contributed to the existence of some lethal compounds in *Euphorbia helioscopia* which may not be exist in *Euphorbia hirta* and *Buxus wallichiana* or may be in minor quantity. It is concluded that further research work on identification of active ingredients of *Euphorbia helioscopia*, *Euphorbia hirta* and *Buxus wallichiana* extracts needed which is more effective than other extracts with low cost and environmental friendly.

## FIRST REPORT OF AN INVASIVE PEST, SPODOPTERA FRUGIPERDA (J E SMITH) (LEPIDOPTERA: NOCTUIDAE) ON CORN IN SOUTHERN SINDH, PAKISTAN

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Invasive species pose always a serious threat to agriculture and cost billions of dollars in terms of reduced production and productivity. The recent preliminary survey in few districts of southern Sindh showed an apprehension of the entry of one more new invasive pest belongs to Genus Spodoptera known as Fall Armyworm (FAW), S. frugiperda (J E Smith). The identification for confirming this alien pest was made based on morphological characters as previously described by Food Agriculture Organisation (FAO) and CABI. The larval samples were collected from corn field of three districts i.e. Hyderabad, Matiari and Tandoallayar. All the collection was brought and further reared at Department of Entomology, Sindh Agriculture University Tando Jam. At the emergence of adults, the males were dissected to study genitalia and compared with pertinent literature. The microscopic study revealed that male genitalia of the collected species had single lobe of coremata which is the main identification of FAW. In addition, the genitalia was with two right and left valves, both broad and quadrate in shape with curved ampulla and pointed at the top. In base of both valves, clavus was short with costal process narrow, straight, elongate and inclined. Juxta was concave at base and with dorsal process. The wing characteristics showed that the males had dark gray and brown shaded mottled forewing with conspicuous triangular white spots at the tip of the wing. The forewings of females were less distinctly marked except few faint grayish brown markings. There were six larval instars of FAW; however in the third larval instar, body became brownish and lateral white lines begun to form. The best identifying feature of the FAW in mature larval stage (5th -6th instar) was a set of four large rectangular spots on the upper surface of the second last segment of body and the face of larvae with obvious mark of inverted white "Y" shape. The head capsule width was ranging 0.3-2.6 mm and the larvae attained length of about 2-33.08 mm (1st-6th instar). The pupa was reddish brown in color, 14-18 mm in length and about 4.5 mm in width. Based on the characteristics those studied in the present study thus confirmed the presence of *S. frugiperda* in Southern Sindh, Pakistan. However, the further identification based on molecular studies is much needed in order to make more confirmation of its corn strain.

### FARMERS' PERCEPTION ON BIOLOGICAL CONTROL OF PAPAYA MEALY BUG IN COASTAL AREAS OF PAKISTAN

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Biological control interventions were taken in 2014 to control an exotic mealy bug species Paracoccus marginatus in papaya at Karachi. Initially survey was conducted for endemic fauna associated with the mealy bug. Then to maximize natural biological control a natural enemies field reservoir (NEFR) was established at Memon Goth where parasitoids and predators developed on debris of mealy bug infested plants that dispersed naturally in the environment. Besides this the parasitized host mummies collected from the debris at NEFR shelter were glued on cards and distributed among farmers at Memon Goth, Darsano Channo, Landhi, Gadap and Kathore. This activity was conducted for two years in 2014-16. To know the farmers' perception of effectiveness of biological control a third-party survey was conducted by Agriculture Extension Department Karachi in 2017. For the purpose a questioner was developed for response from the farmers. As per analysis of this survey with enhanced confidence of farmers on biological control of mealy bug the papaya cultivation at Karachi increased to 210.72 hectares in 2016-17(about 76.51 ha more) compared with 134.21 hectares in year 2014-15. A good difference was seen in papaya production 54015 kgs production in 2016-17, as compared 30040 kgs in 2014-15, estimated output increased by more than half. Before biological control interventions farmers applied 8-10 times sprays in orchards to control mealybug. This frequency of pesticides applications reduced to 1 - 2 sprays at distances of 10 KM from NEFR whereas pesticides sprays completely stopped in 5 km radius of the NEFR. About 97% farmers expressed satisfaction on biological control They had consistent demands for cards of parasitized host mummies to which we gave the name of bio-cards.

# EVALUATION OF ALTERNATE HOST PLANTS OF LEMON BUTTERFLY, *PAPILIO DEMOLEUS* LINNAEUS 1758 (LEPIDOPTERA: PAPILIONIDAE) IN VITRO CONDITIONS AT DISTRICT NAUSHAHRO FEROZE, SINDH

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Lepidoptera is the second largest order than the Coleoptera of class Insecta, which includes moth and butterflies. *P. demoleus* is distributed South-East Asia from Iran across Pakistan, India, China, Japan and Australia and is a key pest of citrus orchards found throughout the year. Intensive infestations caused defoliation of the tree retards the growth of citrus plants and cause economic losses in the form of yield reduction. *P. demoleus* consist of four blackish wings, with white and yellow markings commonly observed with an-association of male and female at citrus plant especially lemon orchards. This research study was carried out on biology, morphology, evaluation of alternate host plants and the biological control of the pest. During present study *P. demoleus* population were evaluated from other citrus fruits including; *Citrus paradisi, Citrus sinensis, Citrus limetta, Citrus reticulata*, and *Citrus aurantium* found maximum pest population during the month of October at the average of 4.79±103°, 4.19±0.84°, 3.51±0.77°, 2.96±0.66°, 2.46±0.37° and minimum in the month of December at 1.27±0.42°,1.12±0.39°, 0.91±0.34°, 0.78±0.28° and 0.54±0.13°, respectively. The natural enemies such as; spiders, coccinellids, praying

mantids, bugs, predators birds and other general predators were observed under field conditions. It was concluded that the larvae of the lemon butterfly pass five stages and vigorously they are harming the citrus plantation. Keeping in mind the above precious ideas and present scientific work will be supportive to the people of Nuashahro Feroze, Sindh-Pakistan. Most of the growers of this area are engaged by lemon plantation as their earning source. But unfortunately, there is a lack of awareness in local formers and severe economic loss occurs due to *P. demoleus* larval infestation.

### POPULATION ABUNDANCE AND REGRESSION ANALYSIS OF DUSKY COTTON BUG, OXYCARENUS LAETUS, KIRBY (HEMIPTERA: LYGAEIDAE) ON BT., COTTON CROP AT DISTRICT KHAIRPUR, SINDH PAKISTAN

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The study over population abundance was carried out at an experimental field of CARS, Kotdiji during, 2018. The trial was laid out in (RCBD) and replicated 3 times. There were fourteen treatments of cotton crop sown with a set of eight Bt., cotton varieties viz., T<sub>1</sub>= CIM-602, T<sub>2</sub>= Bt-109, T<sub>3</sub>= F.H-114, T<sub>4</sub>= F.H-142, T<sub>5</sub>=Bt-121, T<sub>6</sub>= Bt-102, T<sub>7</sub>= Bt-333 and T<sub>8</sub>= CIM-598, respectively. The regression analysis of pest and natural enemies was applied with temperature and relative humidity (RH %). It was observed under the field conditions that the 4th nymphal stage lightly wing pads and 5th stage fully wing pads developed on the thorax of male and female of the pest and female was larger in size than the male. The results indicated that the lower population of DCB was recorded among Bt. varieties viz., (CIM-602, Bt.-333 and Bt.-121) with a mean population (8.74±2.69b, 9.00±2.44b and 9.30±2.63b) per plant was recorded that shows the significant difference among all Bt., varieties. The results of regression analysis indicated that the Dusky cotton bug remained a negative correlation with natural enemies, temperature and humidity. It was concluded that the population of DCB was observed in all varieties of Bt. cotton and no variety was found to be resistant against this pest. It is further suggested that the growers may cultivate the cotton varieties (CIM-602, Bt.-121 and Bt-333) of less responsive to Dusky cotton bug under field conditions and it will be helpful to the growers for understanding the trend of this vigorous pest attacking all over the crop duration and to use the better management practices at the right time. The weather data would be helpful to the growers for making future planning and in reducing the DCB, population management.

### CONTROL STRATEGY OF JASSID, AMRASCA BIGUTTULA BIGUTTULA AND THEIR EFFECT ON COTTON PRODUCTION ON COTTON CROP UNDER FIELD CONDITIONS

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The research study was conducted on the comparison of some synthetic and botanical insecticides with 5 treatments and replicated 4 times in the plot size (2178') on (RCBD) under field condition of the cotton crop. Therefore, the insecticides were applied such as; T<sub>1</sub>= Acephate (Safate) 75 SP, T<sub>2</sub>= Nitenpyram (Amrasca) 10 SL, T<sub>3</sub>= Boiled Neem Leaves, T<sub>4</sub>= Neem Oil and T<sub>5</sub>= Control (un-sprayed) against jassid, *A. biguttula biguttula* at taluka Gambat, district Khairpur - Sindh during, 2018. The data was recorded early in the morning from randomly selected (n=20) plants of each treatment before 24h and after 24h, 48h, 72h, 7<sup>th</sup> day and 12<sup>th</sup> day of each spray. Among the synthetic insecticides the overall results indicated that the Nitenpyram insecticide proved highly effective and reduced the jassid pest population (81.65%), followed by Neem Oil (65.75%), Acephate (64.44%) and Boiled Neem

Leaves (55.20%) however; in control plot least mean population of Jassid was observed (1.75) in all the four sprays. The impact of insecticides on weight of green bolls of cotton was compared; which indicated that in Neem Oil used plot the weight of the green boll was (13.87 g), followed by Bioled Neem Oil (12.73 g), Nitenpyram (11.38 g), Acephate (11.28 g) and in control plot (10.51 g). Among the suggested treatments of insecticides, Neem Oil used plot provided the highest yield, 230 kgs, followed by Boiled Neem Leaves 212 kgs, Nitenpyram 178 kgs, Acephate 168 kgs and in control plot 150 kgs. So it was concluded that both; synthetic and botanical insecticides proved to be better in controlling cotton Jassid; while botanical insecticides were found better in increasing green bolls weight and cotton production as compared to synthetic insecticides.

### OCCURRENCE OF PARASITOID, ERETMOCERUS EREMICUS FOR CONTROLLING OF COTTON WHITEFLY, BEMISIA TABACI UNDER FIELD CONDITIONS AT DISTRICT: KHAIRPUR, SINDH

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The experiments were conducted on the population of whitefly on cotton leaves under field and laboratory conditions (kept unsprayed) at Shah Abdul Latif University, Khairpur during, 2017. The data were collected from randomly selected twenty plants per week for the whitefly pest population and its parasitoid emergence that appeared from seedling to crop harvesting. The whitefly population was counted from various portions of the plant (upper, mid and lower region) under field conditions while; the highest population of whitefly was calculated on the leaves of middle region of the plant  $4.45\pm0.53$  in September, and minimum population of whitefly was counted on the leaves of bottom portion of plant  $0.53\pm0.54$  in July, at the same time overall mean population of whiteflies per week was compared among four months of data collection, among these months the maximum per week whitefly population was observed in September  $4.38\pm0.05$ , and the minimum per week whitefly population was calculated in July  $0.72\pm0.14$ . While; during the laboratory conditions the maximum population percent of the parasite, *Eretmocerus eremicus* was calculated in August  $21.43\pm0.89\%$  and minimum parasite % was recorded in July  $19.37\pm0.89\%$ . It is concluded that the whitefly population and the parasite, *Eretmocerus eremicus* population was remained fluctuated throughout the season. It is further suggested to promote the natural enemy, *Eretmocerus eremicus* that may control the vigorous pest as; whitefly of the cotton cultivated crop at an upper region of Sindh province.

# LIFE TABLE SPAN OF LESSER DATE MOTH, *BATRACHEDRA AMYDRAULA* (LEPIDOPTERA: BATRACHEDRIDAE) UNDER AIR-CONDITIONED TEMPERATURE AT DPRI, SALU - KHAIRPUR

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The biological studies of lesser date moth on date palm fruits during summer season were carried out under laboratory conditions on 5 treatments and 5 replications under air-conditioner temperature started from egg to adult of the pest at Date Palm Research Institute, SALU - Khairpur. It was observed that the overall mean population of eggs by a female with the overall mean population of fecundity was (15.33)/ female and fertility (83.14)%. During the larval stages, the first instar stage took (3.40), second (5.20), third (4.80), fourth (6.20) and fifth (7.40) days with the overall mean of days were (27.00). The pupae took (11.26) with the developmental days (38.26) days and the adult longevity of females (8.38) and male (4.70) days. The life span of female was (46.64) and male (45.6) with the sex ratio of male and female (1:2), respectively. The fluctuation was observed in the day consumption in the life cycle. It is further recommended that the larval stage was the most voracious feeder of dates at that time the

management techniques which are safe to live beings should be applied at well known as Queen of dates, district: Khairpur, Sindh.

## BIO-PESTICIDAL POTENTIAL OF CRUDE VENOM OF *CYRTOPHORA CITRICOLA* (SPIDER: BUTHIDAE) AGAINST *BREVICORYNE BRASSICAE* (HEMIPTERA: APHIDIDAE)

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There is much interest in the development of eco-friendly insecticides because chemical insecticides are harmful for environment. The venoms of arthropod predators such as spiders are rich in compounds that can be used as bio-pesticides. Present study was planned to evaluate the insecticidal potential of different concentration of crude venom of *Cyrtophora citricola* (Spider: Buthidae) against *Brevicoryne brassicae* (Hemiptera:Aphididae). Dissecting method was applied to remove venom glands from spiders. Oral method was used for applying venom on model pest. Venom was applied with concentrations of 0.5ul, 1ul and 1.5ul and the observed %age mortality was 63.3, 93.3 and 100 respectively after 18 hours. The values of LT50 and LT95 are 8.44 and 15.78 respectively. The values of LT50 and LT95 decrease with the increase of venom concentrations. Findings suggested that *C. citricola* can be used in the formation of bio-pesticides.

### COULD EXPLORING OF ENTOMOPHAGY BE AN EFFECTIVE TOOL IN MANAGEMENT OF DESERT LOCUST?

### Ahmed Ali Samejo and Riffat Sultana

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Desert locust is a swarm producing and notorious pest since ancient time. Recently it hit Pakistan in unbelievable figures. During 2019 outbreak and plague desert locust destroyed about 80-100% livelihood and agriculture in Thar (with exception of other regions). In this regard Pakistan expend million liters pesticides to overhauled this sudden natural disaster but unfortunately, we couldn't get proper and effective results against this and still Thari peoples (including others) are in problem. In order to give an alternative remedy, we have attempted to introduce the entomophagy practice amongst the public masses. For this study, Mahandre-jo-par village was selected as a type-locality of Thar Desert. Awareness campaign was launched in this village in order to promote 3 basic themes: 1. Collect the locust (large number) 2. Eat the locust (daily diet) 3. Nutritional value of locust (its protein rich insect). After our demonstration when a huge swarm of about 4-5 km<sup>2</sup> landed on 4 July 2019 near village, villagers immediately attacked on swarm of locust, collected them in sacs and fetched to home for feeding their families. Moreover, boiled and desiccated the locusts then made many crunchy fried snacks. Approximately 60 kg locusts were collected by different groups of villagers every night with the help of hand lights, headlights of vehicles and light traps. Likewise, two other swarms arrived during September 2019 in this vicinity and same practice was followed by villagers again till 3 nights in this result very low population of locust was escapes from this region. Present study recommends publically this campaign would be launch on large scale because if same practice has been adopted in infected areas of locust in other regions hopefully results would be different regarding control. Beside this, locust is not only protein rich insect but it also contains adequate amounts of fat, iodine, phosphorus, iron, thiamine, riboflavin, niacin, as well as traces of calcium, magnesium and selenium which makes them a good candidate for Atkins and Paleo types of diets.

# ABSTRACTS PRESENTED AT THE FIRST VIRTUAL CONGRESS OF ZOOLOGY

(40<sup>th</sup> Pakistan Congress of Zoology – International) December 17-19, 2021

### **SECTION – II**

### PESTS AND PEST CONTROL

## DRYING TECHNIQUES IN FRUITS AND VEGETABLES TO ENHANCE THE SHELF LIFE AND REDUCE THE POSTHARVEST LOSSES

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Drying is known as the best method to preserve fruits and vegetables, decreasing not only the raw material volume but also its weight. This results in cheaper transportation and increments the product shelf life, limiting the food waste. Drying involves the application of energy in order to vaporize and mobilize the moisture content within the porous products. During this process, the heat and mass transfer occurs simultaneously. The quality of dehydrated fruits, vegetables, and aromatic herbs is a key problem closely related to the development and optimization of novel drying techniques. This study reports the weaknesses of common drying methods applied for fruits, vegetables, and the possible options to improve the quality of dried products using different drying techniques or their combination. The quality parameters under study include color, bulk density, porosity, shrinkage, phytochemicals, antioxidant capacity, sugars, proteins, volatile compounds, and sensory attributes. In general, drying leads to reduction in all studied parameters. However, the behavior of each plant material is different. On the whole, the optimal drying technique is different for each of the materials studied and specific conditions must be recommended after a proper evaluation of the drying protocols. However, a novel or combined technique must assure a high quality of dried products. Furthermore, the term quality must englobe the energy efficiency and the environmental impact leading to production of sustainable dried products.

# EVALUATION OF RELATIVE SUSCEPTIBILITY OF *CALLOSOBRUCHUS MACULATUS* (F.) (COLEOPTERA: CHRYSOMELIDAE) ON TWO DIFFERENT STORED PULSES

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The *Callosobruchus maculatus* commonly known as Cowpea weevil is a worldwide insect pest that infests pulses in the fields and seeds in storage. An investigation has been conducted on the life history of Cowpea weevil reared on Green gram and Mash gram pulses through three successive generations. The result obtained revealed that Green gram was susceptible while Mash gram had moderately resistant stored pulses against *Callosobruchus maculatus*.

# POPULATION DIVERSITY OF CHEWING LICE (PHTHIRAPTERA: AMBLYCERA AND ISCHNOCERA) INFESTING DUCKS AND GEESE (AVES: ANSERIFORMES: ANATIDAE) OF SINDH, PAKISTAN

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In Pakistan there are 37 species of ducks and geese representing 23 species in Sindh. Ducks and geese are aquatic game birds and migrate to Pakistan during winter seasons and stay from October to March of every year at various waterbodies in Sindh region. These birds carries both types of parasites ectoparasites as well as endoparasites. The

present investigation of ectoparasites has been undertaken on total 76 birds, including 8 species of ducks and geese, Anas crecca (Linnaeus), Anas clypeata (Linnaeus), Anas platyrhynchos (Linnaeus), Anas querquedula (Linnaeus), Aythya ferina (Linnaeus), Aythya fuligula (Linnaeus), Anser anser (Linnaeus) and Anser albifrons (Linnaeus) were collected from different districts of Sindh Province. Out of 76 birds, only 57 birds were found infested with the prevalence of 75%. Presently, total of 575 chewing lice specimens were being recovered, including six species, Anaticola crassicornis (Scopoli, 1763), Anaticola mergiserrati (De Geer, 1778), Anatoecous icterodes (Nitzsch, 1818), Anatoecous dentatus (Scopoli, 763), Trinoton gurequdulae (Linnaeus, 1758) and Holomenopon leucoxanthum (Burmiester, 1838). The specieswise burden of chewing lice has been recorded maximum by Anaticola crassicornis 170 (29.56%) followed by Trinoton qurequdulae 165 (28.69%), Holomenopon laucoxanthum 80 (13.91%), while minimum burden was recorded by Anaticola mergiserrati 40 (6.95%), Anatoecous dentatus 45 (7.82%) and Anatoecous icterodes 75,(13.04%). The district wise prevalence was calculated, with maximum in Karachi and Badin (100%), then it is also high in Hyderabad (80%), Larkana (76.47%), Jamshoro (71%) and Kambar Shadadkot (70%), whereas the minimum prevalence of chewing lice was found in district Noushahro feroze (57.14%) and Dadu (66.66%). The total prevalence of chewing lice on ducks and geese species was calculated as maximum was recorded in Anas crecca (92.85%), Anas clypeata (87.5%), Aythya ferina (80%), Anas querquedula (75%) and Aythya fuligula (75%), whereas minimum prevalence was recorded in Anser albifrons (55.55%), Anser anser (60%) and Anas platyrhynchos (66.66%). The complete life cycle of these six species were observed on anatid birds their quantitative parameters like prevalence, population abundance, physical effects on feather and seasonal variation of all species of chewing lice were studied in the present project.

### CURRENT OUTBREAK OF SPODOPTERA FRUGIPERDA (FALL ARMYWORM) ON FODDER CROPS (MAIZE) OF SINDH

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The fall armyworm (FAW) Spodoptera frugiperda (Smith) (Lepidoptera: Noctuidae) is an obtrusive polyphagous lepidopterian pest of different yields that cause weighty infestation on vegetables, organic products cotton crops maize, and other fodder crops, especially harming maize and corn, although it is endemic to the American continents, it has spread to Africa and Asia. However, there has been no confirmed report of its presence from Pakistan. Except for the upper Sindh districts of Jacobabad, Larkana, and Shikarpur, the prevalence of FAW has been documented in all maize-growing areas of Sindh. The current study was carried out on fodder crops, specifically maize crops, from July 2020 to October 2021. Fodder maize experienced more damage than other fodder crops. Fall armyworm (FAW) infestation in fodder maize led to the destruction of thousands of hectares of maize. In the Matyari district, extensive damage to fodder maize was observed. The major cause of the current outbreak was the favorable climatic circumstances for numerous invading pests, such as high humidity and moderate temperatures. The current study revealed that climatic change is a significant factor and also a big challenge for governments, academia, and the entire globe. As a result, a country-wide study, as well as molecular identification of FAW, should be carried out to confirm its presence in Pakistan's maize areas. With proper management, this could be helpful for preventing its spread.

#### ABUNDANCE OF MYZUS PERSICAE IN JACOBABAD DISTRICT SINDH PAKISTAN

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Aphids are harmful pests. Aphids not only harm crops but it creates very dangerous diseases. The survey was conducted in January 2017 till February specimens were brought to the insectary of the zoology department at the University of Sindh Pakistan. The identification of aphids was done by standard taxonomic keys. Four Localities of Jacobabad Gahri sabhayo, Dasti, Ali pur and rind wahi were observed. Myzus persicae was found abundantly in months of January and February.

### THE REFLECTION AND ABUNDANCE OF FRUIT FLIES (BACTROCERA SPP.) ON SOME MANGO VARIETIES IN SINDH PAKISTAN

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The fruit fly is one of budding pest that is very harmful to mango production in Pakistan. The originality about fruit fly species in certain area is crucial to scrutinize, pest risk assessment, to develop of appropriate standards for plant quarantine treatment, and to manage of fruit fly monitoring and control programs. This study aims to analyze the fruit fly abundance and composition among three mango varieties ("Chunsa", "Sindhri", and "Beganpali") in Mirpukhas, Hyderabad and Tado Muhammad khan Districts, in Sindh Pakistan. The Infested mangoes were collected from orchards (fields), markets and traditional vendors from above areas. All samples were transferred and reared in the laboratory, larvae to pupae and allowed to emerge as in mangoes. The results showed that *Bactrocera dorsalis* (Hendel), *Bactrocera carambolae* Drew & Hancock, and an interspecific hybrid of both species were found on mangoes in above Regence. The *Bactrocera zonata* was merely found on mangoes in same areas. The *B. dorsalis* was the abundant species in all mango varieties. There is (+) correlation between the nutritional content of mangoes with the fruit flies species diversity and pupae weights of fruit flies.

### SURVEY OF ORTHOPTERA PESTS AND THEIR AFFLICTION WITH PLANTS FROM VARIOUS AGRICULTURAL REGIONS OF PAKISTAN

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A preliminary survey was conducted in various localities and important pests of 3 families were observed to cause huge damages in valued crops i-e *Phyllo choreiara makrishnai* (Eumastacoidea) makes leaf like appearance, stick grasshoppers (Proscopiidae) do mimicry like wooden sticks in form and coloration. They also use advanced legs for escaping and jumping. They are considered as destructive pests because it destroys most economical crops, their outbreak seen in hot and dry weather, they also damage fruits and woods. Beside this few species of family Tettigonidea was also seen. Most of their activity in the field has been observed in daytime but they are highly active at nighttime and produce different sounds. They spend the whole day in vegetation and at night they engage in reproductive activities. In order to compile all the orthoptera pests a detailed survey is needed.

# INSECTICIDAL POTENTIAL OF INDIGENOUS WEEDS EXTRACTS OF BAHAWALPUR PAKISTAN AGAINST DUSKY COTTON BUG (OXYCARENUS HYALINIPENNIS) (HEMIPTERA: LYGAIDAE) UNDER LABORATORY CONDITIONS

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This study evaluated the aqueous extracts of three weeds namely *Datura alba*, *Chenopodium album and Withania somnifera* for insecticidal potential against *Oxycarenus hyalinipennis* (Hemiptera: Lygaeidae) under laboratory

conditions. Extracts were prepared from three plant parts *e.g.*, leaves, stems and roots and applied at six concentrations ranging from 1 % to 20 % and a control treatment with water only. Mortality of pest insects was checked after three exposure periods *e.g.*, 24 hours, 48 hours and 72 hours. Results showed maximum mortality was generated due to 20 % concentration of extracts. Roots extracts were more potent followed by leaves and minimum mortality was due to stem extracts. Maximum mortality of insects was after 72 hours period followed by 48 hours and minimum was after 24 hours period. Among the three weeds *D. alba* proved more toxic in terms of mortality of *O. hyalinipennis* compared with other two weeds. These results pinpoint towards *D. alba* weed with potential to evaluate further for insecticidal properties for different important sucking insect pests of agricultural crops including *O. hyalinipennis*.

### INFESTATION OF *LIPOSCELIS ENTOMOPHILIA* (PSOCOPTERA: LIPOSCELIDIDAE) ON ORTHROPTERA PESTS

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Serious infestation of *Liposcelis entomophila* (Enderlein) (Psocoptera: Liposcelididae) was observed in the insects preserved in Sindh Entomological Museum. *Liposcelis entomophila* is a nuisance pest that seriously threatens the safety of stored products. During the museum survey 15 boxes were found with heavy infestation on 242 specimens. All the insects preserved in these boxes have great economic value in evolutionary linkage. All the stages of *L. entomophila i-e* eggs, early-instar nymphs, and adults were seen in insects' bodies. It has been demonstrated that the survival of Liposcelida is highly dependent on the atmospheric moisture level.

### IN SILICO ANALYSIS OF REPELLENT ACTIVITY OF DIMESTEROL AND CHAVICOL AGAINST AEDES AEGYPTI

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Dengue is a mosquito-borne viral disease that is spread by vector *Aedes aegypti*. This disease has a global incidence of 50 to 400 million per year and is epidemic in Pakistan. Mosquito repellents are necessary to control mosquito borne diseases as these diseases poses a national and world-wide health and economic risk. However, the use of synthetic mosquito repellents has been reported to be associated with various environmental and health issues. Previous study from our institute has reported the potent repellent activity of the organic extract of *Illicium verum* against the *A. aegypti*. The GC-MS of the extract revealed the presence of trans anethole, chavicol, estragole and dimestrol as its major components. In the present project effectiveness of chavicol and dimestrol as a mosquito repellent was evaluated against target protein AaegOBP1 using in silico techniques. MCULE software was used for docking of ligand to protein. Results were visualized using UCSF chimera and LigPlot+. Docking results of understudy compounds were compared with docking results of DEET, that exhibited H-bonding with Met91 residue of binding pocket and binding energy ΔG-7.7 kcal/mol. Dimestrol showed simmilar binding parameters with AaegOBP1 with free energy ΔG-8.0 kcal/mol and a H-bond with Ile125 residue of the binding pocket. Chavicol also proved to be a good competitor of DEET with binding energy ΔG-6.8 kcal/mol and H-bonding with Phe123. Dimestrol and chavicol were found to be safe compounds to use as topical repellents using ADMET analysis. In-vitro repellent bioassays should be performed to validate the efficacy of dimestrol and chavicol to be used as repellent.

### EFFICACY OF ENTOMOPATHOGENIC FUNGI AGAINST COTTON MEALY BUG PHENACCOCUS SOLENOPSIS TINSLEY PSEUDOCOCCIDAE: HOMOPTERA

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Cotton, Gossypium hirsutum is a cash crop. Cotton crop is attacked by many pests and all of those pests cause severe damage on cotton crop and cause 20% to 40% yield loss every year. Firstly, mealy bug was introduced in North America after that in Asia and Europe. Cotton mealy bug causes yellowing, malformation and weakening of leaves. Many insecticides like carbamates, organophosphate and pyrethroids were applied but they give short term control. In case of repeated application pesticide resistance occurred. According to IPM, biological agents are used against insects in the form of entomopathogenic microorganisms. Beauveria bassiana, Metarhizium anisopliae and Isaria fumosorosea are used as entomopathogenic fungi against cotton mealy bug. Biological control agents were cultured in laboratories. Entomopathogenic fungi inoculated in the mealy bug. Among all biological agents, Beauveria bassiana and Metarhizium anisopliae are highly effective on 2<sup>nd</sup> instar of mealy bug with high mortality rate.

### OVIPOSITION PREFERENCES OF PEACH FRUIT FLY (BACTROCERA ZONATA) ON DIFFERENT FRUIT HOSTS

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Bactrocera zonata is a quarantine pest worldwide and affect a variety of fruits and vegetables. The current study was done to investigate the host selection of B. zonata under the laboratory conditions. For this purpose, four different fruits i.e., banana, guava, apple and citrus were used in both choice and no-choice test condition. Result revealed that banana have received the highest oviposition and number of pupal populations followed by guava apple and citrus in no-choice test. The same trend was observed in choice test, the highest pupal population of fruit fly was observed in banana under the choice condition. Furthermore, the lowest pupal population was observed in guava and apple and citrus have no pupal population in choice test. B. zonata responds differentially to co-existing host species in the field under choice and no choice tests, according to the current research. The pest control tactics for fruit crops are influenced by B. zonata's host and choice preferences.

### BOTANICAL INSECTICIDES ON THE LONGEVITY OF ADULT GREEN LACEWING (CHRYSOPERLA CARNEA)

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Green lace wing is natural enemies of numerous insect pests of agriculture crops. To keep its population in field we need to alternate of insecticides because insecticides not kill the harmful insects but also kill its natural enemies. Botanicals pesticides are less harmful for natural enemies but we need further research to confirm the botanicals plant extract influence of green lace wing. So present study was designed to check the two botanicals plant extract Garlic and eucalyptus plants extract influence on adult's longevity of green lace wing. Three different concentration (10%, 20%, 30%) of both botanicals plant was test in artificial diet of green lace wing. In eucalyptus a statistically significantly increased adult longevity was observed in control group, while there was no statistically significant difference was found among 10, 20 and 30 % of treatments. While in garlic a statistically significant enhance adult longevity was observed in control

group, as compared with given treatments, while no significant difference was observed between 10% and 30 % and also 30% and 20 % at adult longevity. But in Both botanicals plant extract was compared regarding adult longevity we observed eucalyptus plant extract were found more effectively compared to garlic extract in all give concentrations. We observed that eucalyptus plant extract significantly reduce the adult longevity of green lace wing as compared to garlic extract in all given concentration treatments. Present data will be helpful for research farmer and scientific community.

# USE OF DIFFERENT COLOURS OF STICKY TRAPS ON THE POPULATION OF WHITEFLY (BEMISIA TABACI)

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White fly is one of the most serious insect pest attacking numerous agriculture crops in Pakistan. A wide range toxic pesticides have been used to mitigate the pest population. However, due to environmental contamination and public health risk the use of toxic pesticides need be minimized. In present study, various colours of sticky traps were used to evaluate population of white fly. Various colours of sticky traps such as Yellow, Red, Green, Blue and Black were used in the field of brinjal crop and thus data was recorded on daily basis for one week. Maximum population of white fly was recorded in Yellow sticky trap (88.99) followed by Green (14.55), Black (11.99) and Red (9.22) colour sticky trap. While a minimum population of white fly was observed in blue trap (7.99). Statistically significantly increased population of white fly was observed in yellow trap and green tarp. While no significant difference was found between black and red traps and also in red and blue traps. It was concluded that yellow color could be used in the field to suppress the population of white fly. The present data will be helpful for the research and scientific community for further research.

### SPLAT-PBW: AN ENVIRONMENTAL-FRIENDLY, COSTABLE MATING DISRUPTION TOOL FOR THE MANAGEMENT OF PINK BOLLWORM ON COTTON

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The farmers of cotton crop have reaped the benefits of Bt cotton cultivation since commercialization [March 2002]. From 2014 the pink bollworm (PBW) outbreak in the larged cotton-growing areas of the country was witnessed. The pest increased and became more problematic for farmers and country economic. Its being controlled with the new management practices due to resistance to cry-toxins and chemicals. A non-chemical approach modifying the behavior of PBW was evaluated its effectiveness in comparison with existing management strategies. An area-wide management trial with disruption mating technology was carried out using pheromone, and lure application technology for pbw. Application of 1250 g/acre of the lure during 2017 in 154 acres and 206 acres during 2018 in India, recorded significant control of PBW. The results that SPLAT-PBW applied at maximum acres was found to be optimum, as minimum rosette flower (7.23%), green boll damage (8.30%),locule damage (7.40%), and higher yield (33.50q/ha) recorded. As compared to farmers' practice which yielded 22 q/ha even after applying the 6 rounds of chemicals spray. At the end of the 5 weeks, 42 of the active ingredient of pheromone was present in the field sample. It's show the slow release pheromone as compares to other. Non-chemical practices of insect pests in the cotton significantly benefit in decreased the load of pesticides and total output. In future insecticidal transgenic crops will use modified Bt toxins and new ways to such as RNA interference(RNAi).

#### OUTBREAKS OF SCHISTOCERCA GREGARIA IN VARIOUS DISTRICTS OF SINDH

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The present study focuses on *Schistocerca gregaria* commonly known as Desert Locust belonging to (Acrididae: Orthoptera) is an economically disastrous pest of the extensive range of crops including cereals. A survey for the collection of the specimens was carried out during the year 2019 from different districts of Sindh, which were recently affected by the swarm of *Schistocerca gregaria* Desert Locusts. The number of collected specimens is 935. Besides this, an immature identification key was also given for paper analysis. This species is observed as polyphagous and can strongly harm a wide range of grasses including wheat, cotton guar, maize, and fruits. This outbreak was the worst in history.

#### THE MANAGEMENT OF INSECT PESTS BY USING ENTOMOPATHOGENIC FUNGI

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Insect pests cause huge damage to our crops. Synthetic pesticides are broadly used for controlof insect pests but this approach has various negative impacts on environment and non-target organisms. Due to excessive use of pesticide over the years the insects developed resistant against these chemicals. So, there is need for alternate option to manage these pests. Among different IPM program, use of microbial formulations is ecofriendly and safe for life. A biological control agent using entomopathogenic fungi has been developed in place of chemical pesticide. Entomopathogens due to their eco friendliness are preferred to control insect pests by contact mode of action. Insect pathogenic fungi have resulted various improvements in mycoinsecticide products based on *Paecilomycesfumosoroseus Beauveria bassiana*. The most effective entomopathogenic fungi included *Metarhiziumanisopliae*, *Beauveria bassiana*, *Cordyceps javanica*. These products of entomopathogenic fungi have been very effective; however, utilization of these products is very low, due to low performance under challenging conditions and lack of awareness. Vigorous research work needs to be done to improvetheir performance under challenging conditions, mass production, formulation, pathogen virulence and spectrum of action. It should be possible that best use of entomopathogens at wider range can eliminate the chemical use at larger level.

# EFFECT OF DIFFERENT PLANT EXTRACTS AGAINST COTTON JASSID NYMPHS, AMRASCA BIGUTTULA ON COTTON

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Cotton is an important fibrous crop. It is one of the major crops of Pakistan. Cotton production play about 10 percent role in national GDP of Pakistan. Cotton is facing a serious threat from sucking and chewing insects for its revival in Pakistan. There are many sucking insect pests of cotton, jassid is one of them. Various synthetic insecticides are used by cotton growers to keep the insect's population below ETL level. Insecticides are expensive, harmful to environment and disturbing the natural fauna as well. Botanical extracts are very useful to manage pests' population under control and are eco-friendly. The present study carried out about to predict efficacy of different plant extracts against nymph of cotton jassid. During study four different plants bio extracts i.e., tobacco (*Nicotiana tabacium*), neem (*Azadirechtin indica*), kortumba (*Citrullus colocynthis*) and heing (*Ferulaassa-foetida*) were sprayed with 15 days of intervals. The result showed that the highest mortality by the tobacco (66.6%), by neem (33.3%), by kortumba (50%) and by Heing (55.6%). It was observed that botanicals were effective until 48 hours which was represented the much

effectiveness of different plant extracts against cotton jassid. This present study suggested that the use of plant extracts is very useful to reduce jassids population without harming the natural enemies.

# EFFECT OF VARIOUS CONCENTRATIONS OF PYPROXIFEN AND LARVIN ON AEDES EAGYPTI EGGS

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The experiment was carried out to record the effect of Effect of various concentrations of Pyproxifen and larvin on Aedes eagypti eggs. Aedes aegypti (Diptera: Culicidae) is the main mosquito responsible for the transmission of dengue in tropical and subtropical regions of the world. Presently no vaccine is available for the prevention of dengue virus infection at the world level. Therefore, control of dengue vector is the only way of dengue management. The present study was carried out at the Nuclear Institute for Food and Agriculture (NIFA), Peshawar in Plant Protection Division during November, 2017. Two different insect growth regulators (IGR) as Pyriproxyfen and larvain were applied against freshly laid eggs of Ae. aegypti at six different concentrations as 2,4,8,16,20 and 40 ppm. Each concentration was repeated three times. The control was treated only with tap water. The counted no of freshly laid eggs of Aedes aegypti were exposed to all the tested concentrations under laboratory conditions (27°C, 75±5 RH). In the overall results, found the significant results in the context of eggs inhibitions/ hatching in IGR (50.240% 49.731%) and larvain as (62.291%, 37.700%) respectively. Similarly among the tested concentrations, significantly recorded the highest level (40ppm) was highly effective followed by 20, 16,8, 4 and 2ppm) for maximum eggs inhibition. In the control, maximum eggs hatching was found as (81.867% 88.160) respectively with IGR and Larvan treatment. It was concluded that IGRs can be utilized as environment friendly control measures for Culex quinquefasciatus and Aedes albopictus spp of mosquitoes on small and large scale. This will reduce the use of conventional insecticides by the public health authorities and help in reducing selection pressure of insecticides.

# POPULATION FLUCTUATIONS OF FRUIT FLIES, BACTROCERA SPP. IN MANGO ORCHARD ECOSYSTEM OF SINDH, PAKISTAN

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Keeping in view the economic importance of fruit flies and losses incurred by *Bactrocera spp*. infestation which caused ban on export of mango to Europe. Studies on population fluctuations of Peach fruit fly *Bactrocera zonata* (Saunders) and Oriental Fruit Fly *Bactrocera dorsalis* (Hendel) were undertaken during 2018 on mango fruit in different climatic zones of Sindh Province. Investigations were carried out at district Larkana and district Hyderabad in mango orchards using methyl eugenol lured traps. The male lure tempted traps were displayed at three meter height and replenished on fortnightly basis in experimentation sites. Results revealed that significantly highest (658.6±20.26, 601.9±25.38) population of *B. zonata* were recorded in the month of June in mango orchards of district Hyderabad and Larkana. Similarly, higher (447.4±18.3, 396.9±38.79) population of *B. dorsalis* was recorded in June in Larkana and Hyderabad regions. Moreover, lowest (7.7±0.96, 5.9±0.97) population of both species were recorded in the month of December in Larkana. On contrary lower (12.9±1.35, 9.5±0.45) population of *B. zonata* and *B. dorsalis* were observed in the month of January in mango orchard eco-system of Hyderabad region. The ensnaring of both species were positively correlated with the temperature while relative humidity has slight negative effect on it. Results of the present investigation would be helpful in developing sustainable male annihilation technique (MAT) program as an important IPM component for various mango orchards in Sindh.factors, MAT.

# INVASION OF FALL ARMYWORM (FAW), SPODOPTERA FRUGIPERDA FROM MAIZE AND ITS SURROUNDING AGROECOSYSTEM IN HYDERABAD

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The fall armyworm (FAW), Spodoptera frugiperda is a sporadic pest that belongs to a Lepidopteran family Noctuidae. At present time, It is a rising threat to food and nutritional security of millions of people throughout the world as its voraciously feeds on more than 80 plant species including many crops causing major damage to economically important cultivated grasses particularly corn, sorghum, rice, sugarcane, wheat and other vegetable crops and cotton etc. It can cause famine in areas where cereals are subsistence crops. It is native to America but due to its potent flying abilities since the first detection of invasion, it rapidly spread to West Africa and to throughout sub-Saharan Africa and now it has been reported in most of the Asian countries like China, Myanmar, Thailand, India, Bangladesh and Sri Lanka. Pakistan is not an exception as its presence has also reported from Faisalabad and some localities of Sindh, Pakistan. Different biological characteristics like wide host range, inherent ability to survive in a broad range of habitats, strong migration capacity, high fecundity, rapid development of resistance to insecticides contribute to FAW invasiveness.

### SECTION – I I I

### ENTOMOLOGY

### RELATIVE ABUNDANCE OF ORDER ODONATA, HYMENOPTERA AND HEMIPTERA IN RICE FIELDS

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Rice (Oryza sativa) is important cereal crop in world and also serves as the primary source of staple food for majority of global population. Insects possess very important rank in rice fields owing to their role in pollination, decomposition, herbivory and biological control. They are also indicator of biodiversity by monitoring the changes in natural habitats resulting disturbance in local diversity. The present study was designed to measure the diversity of order Odonata, Hymenoptera and Hemiptera along with other related aspects in rice fields. Specimens pertaining to these orders were collected from rice fields of District Faisalabad by using hand picking, hand net and forceps. A wide range of diverse fauna was collected from this field. The whole sampling was done randomly from October 2013 to April 2014. Collected insects were identified on the basis of their morphological characteristics with the help of taxonomic data and various aspects of faunal diversity. A total of 290 specimens were collected from this crop pertaining to order Odonata, Hymenoptera and Hemiptera. Maximum relative abundance (6.21%, N=18) was recorded for Cassida rubiginosa (Hemiptera: Chrysomelidae). However, least relative abundance was recorded in case of Ischnura spp. (Odonata: Lestidae), Euchistus servus (Hemiptera: Pentatomidae), Triatoma infestans (Hemiptera: Reduviidae), Leptocorisa acuta (Hemiptera: Alydidae), Nabis kinbergii (Hemiptera: Mirridae) and Macrocentrus grandii (Hymenoptera: Braconidae). As for as relative abundance is concerned up to family level, in rice fields, among total of 290, it was recorded maximum for family Formicidae, Pentatomidae, Alydidae, Lygaeidae and Delphacidae ( $N \ge 20$ ). Highest relative abundance of Order Hemiptera was recorded in rice fields as compared to Odonata and Hymenoptera. Relative abundance was recorded as 29.02% in rice field. Diversity (H'), Evenness (J) and Dominance (D) was recorded as 2.2707, 0.9221 and 0.0779, respectively.

### EVALUATION OF INSECT PESTS (CHEWING BOLLWORMS) INFESTING BT AND NON BT COTTON CULTIVARS

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Transgenic organisms are becoming a fashion for modern world. Current study is focused on performance of transgenic cotton. Chewing bollworms infest cotton crop and lead to massive damage. In this study we investigated the influence of pests on Bt and Non Bt cotton. Results showed that non Bt varieties are more influenced by chewing worms as compared to Bt variety. We found that environmental stresses bring the changes in the affectivity of Bt cotton variety against chewing worm. Before this, no work was done to study the impact of chewing worms on Bt and Non Bt variety in this area. The objective of the present study is to investigate the influence of chewing pests on Bt and Non Bt cotton varieties in Southern Punjab (Pakistan). For this purpose different cotton variety i.e. Niab Non Bt and four Bt varieties viz. IUB\_33, MNH\_988, Lalazar and Sitara were grown in four regions (Multan, Khanewal, Muzaffar Garh and Kabirwala). These cotton varieties were surveyed for the collection of data on different pests (Army, Pink and Spotted bollworms) populations and eggs counts from April 2014 to October 2016. It was found

that Niab Non Bt always borne the highest number of chewing worms and their eggs as compared to Bt varieties. Bt cotton varieties showed significantly higher resistance than Non Bt variety against chewing bollworms of cotton as indicated by the presence of lesser number of adults and eggs of three types of boll worms on these varieties under climatic conditions of southern Punjab in Pakistan. Non Bt cotton varieties had poor HPSI as compared to Bt varieties throughout the study period at all four sites. It is therefore suggested to promote Bt cotton varieties to reduce economic losses borne by cotton farmers as Bt cotton varieties possess greater resistance against pests specifically chewing bollworms.

#### EVALUATION OF CUE-LURE PHEROMONE FOR THE ATTRACTION OF MELON FRUIT FLY BACTROCERA CUCURBITAE IN BITTER GOURD VEGETABLE CROP

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Fruit flies constitute the most important group of pest infesting fruits and cucurbitaceous vegetable crops throughout the universe. Particularly Melon fruit fly has been weighed notorious pest. For the management of this pest, various control measures have been advocated in all over the world. During the performed research work assess the Persistency and Durability of cue-lure sex pheromone and of three different insecticides including Limda, Amida, and Route. Keeping in view sex pheromone was used for the attraction as well trapping of flies under laboratory conditions and the insecticides mixed with cue-lure for the killing of pest inside the traps was observed as triplicate under field conditions. The four different persistency durations such as 15, 25, 35, 45 days were observed in bitter gourd crop. Results revealed that significant higher *B. cucurbitae* were captured in the fifteen days of replenished which (41.89±1.83a). However, significantly lower (19.44±1.46e) after sixty days of replenished. It was observed that lure might be replenished after every fortnight in the month of May, June and July-2016. Moreover, results also depicted that considerably higher (49.62±1.95) killing of *B. cucurbitae* were observed in route and lower (38.30±1.92) in Amida. The results of the study would be helpful for the selecting of the insecticide in mixing with cue-lure for the management of the cucurbit flies under field conditions.

#### DISTRIBUTION PATTERNS IN ORTHOPTERA BIODIVERSITY FROM CHOLISTAN DESERT

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The Orthoptera have inhabited the Earth for ca 300 million years and today include about 24,000 known species but it is estimated that this figure may double when extensive survey were conducted in different Toba of Cholistan desert at regular base. During present survey fair numbers of terrestrial insects commonly known as short-horned grasshoppers, katydids, bush crickets, crickets, and locusts were collected. Its morphology and morphometry was discussed along with its effect on some medicinal plants in desert. We examined that species can be diurnal (all most short-horned) and nocturnal (long-honored) were maximum in numbers in field. We have studied a series of facts about orthopteran systematic and their enormous biodiversity, concentrating on ecological and evolutionary characteristics such as habitat variation, modes of feeding, along with documentation of more direct human interest as their use as food their importance in folk medicine and their role as entertainment was also given.

# SYSTEMATIC AND MORPHOLOGICAL STUDY OF THE GENUS *AIOLOPUS* FIEBER (ACRIDIDAE: ACRIDOIDEA: ORTHOPTERA) WITH SPECIAL REFERENCE TO ITS PHALLIC COMPLEX

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At the present orthopteran fauna in agricultural fields of Pakistan were investigated. Previously large numbers +of specimens have been collected from different provinces of Pakistan. Three species namely *Aiolopus thalassinus thalassinus*, Fabricius, A. thalassinus tumulus, Fabricius, and A. simulatrix simulatrix, Walker, of subfamily Oedipodinae were studied. However, the most dominant and widely distributed species was Aiolopus thalassinus thalassinus its distribution has been sorted throughout country. Some important characters of male and female genitalia have been described. All these species were recognized as severe pest of many important crops in Pakistan. Aiolopus spp. are widely distributed from agriculture range land to semi deserted regions in country. In the current study attention was paid to recognize the male and female genitalia characters; that play an important role for taxonomic purpose and to solve the problem of identification. This genus is closely related to Platypygius but has significant difference in it phallic complex.

# VARIATION IN THE SPERMATHECA OF *OXYA* SPECIES (OXYINAE: ACRIDIDAE: ORTHOPTERA)

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A comparative study on the spermatheca of Oxya species has been carried out. An attempt has been made to describe and illustrate different structures of spermatheca of female in five Oxya species viz: Oxya hyla hyla Serville, 1831, O. velox (Fabricius, 1787) and O. fuscovittata (Marschall, 1836), Oxya nitidula (Walker 1870) and Oxya kashmorensis. The spermatheca of Oxya species consist of a ductus seminalis and a receptaculum seminis which ends in two blind sacs which are called the pre apical and apical diverticula. The diverticula of Oxya species show significant morphological variation that might be implicatetheir functional variances. A microscopic investigation of the spermatheca showed that the presence of numerous gland ductules, but a lack of acanthae, which is mostly present in many acridids. The purpose of this study to discover their significance in order to make the perfect and authentic identification of genus and species. Genitalic structures particularly female spermatheca makes it possible to put onward some suggestions regarding interrelations of family Oxyinae more clearly than the external characters.

# ECOLOGICAL DISTRIBUTION AND SEASONALITY OF DARKLING BEETLES (COLEOPTERA: TENEBRIONIDAE) FROM LOWER SINDH

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The survey of Tenebrionidae (Darkling beetles) was conducted to establish species knowledge to assess habitat and diversity of different family of Darkling beetles around various region of lower Sindh. The specific invertebrate species diet in this region is poorly known and this study helps to understand those needs for attempts restore populations in the different areas of Sindh. Pitfall traps were used to study the diversity and seasonal abundance of invertebrates, especially sub family Stinochinae, Tenebrioninae and Pimeliinae were reported four cities of lower Sindh i.e. Hyderabad, Tando Jam, Badin, Jamshoro. From March 2018 to September 2019. Traps were checked monthly and analysis was done based on the total catch of Tenebrionids from all study sites. The most abundant were Tenebrionids specimens. From all sites a total of 600 Tenebrionid beetles were trapped, representing 04 species i.e.

Promethis semisculcata (Fairmaire, 1882), Strongylium forcipicolles (Fairmaire, 1900), Trachyderma hispida (Forskal, 1775), Pimelia capito (Krynicky 1832). The most dominant were Trachyderma hispida and Pimelia capito. The species Promethis semisculcata was only found in Jamshoro site while species Strongylium forcipicolles was in Badin site. Number of Tenebrionids trapped in winter and midsummer were low, they reached peak in the spring when the weather was moderate and plant diversity highest. The appearance and disappearance of the beetles were strongly linked with seasonal changes.

# INFESTATION OF *HIEROGLYPHUS ORYZIVORUS* (HEMIACRDIADANE: ACRIDIDAE: ORTHOPTERA) ON DIFFERENT CROPS VARIETIES IN KASHMORE DISTRICT

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The *Hieroglyphus oryzivorus* is reported as a major pest of rice, sugarcane, wheat, maize and as a minor pest of millets and fodder crops in Sindh, Pakistan. During the present investigation, a total of 1026 specimens were collected from Buxapur and Badani a major crop growing areas of Kashmore. The investigation was carried from January to December. Both Macropterous and Brachypterous forms of *H.oryzivorus* were collected from important varieties of rice i-e Aery (Sindhi) was infested 12.78% in 2016 while Hybrid (Dhaga) showed highest infestation ratio of 44.4% while, Pukhraj (Hybrid rice) was infested by 4.%, Royal (Hybrid rice) was 7.07%, Sattar (wheat variety) was infested 2.94%, TD (wheat variety) was infested by 4.91, Torhy (sindhi wheat variety) was infested 1.67%, Onion was infested by 2.94% in 2016 while, Bringal was infested by 5.11%, leady finger was infested by 3.99%, pea was infested by 9.24% and surprisingly gram was no infested the same case was seen in cucumbers. The above data shows that insect is invading to new habitat and making adaptation in different season which is alarming condition for future swarm. Multiple cultivation of Dhaga (Hybrid rice variety provide the ideal breeding place for enhancement of this pest population. During present survey it was noted that the cold month like January) and very dry month (May) the species do not survive. The rain fall was the major abiotic factor for hatching of that species.

# SEASONAL OCCURRENCE OF BOLLWORMS IN COTTON FIELD FROM DISTRICT GHOTKI, SINDH

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The present survey was conducted in cotton growing areas (Ghotki, Khangarh, Mirpur Mathelo, Dharaki, Ubauro) in district Ghotki. Extensive survey was conducted in order to determine incidence, prevalence and distribution of major cotton bollworms (pink bollworm, spotted bollworm and spiny bollworm). At least 15 random and 15 non-random samples was collected from each area in farmers' field by moving diagonally in the field and samples were collected from available fresh ratoon crop. The findings of the present survey shows that in taluka Ghotki maximum population of pink, spotted and spiny bollworms (69, 24 and 18 nymphs) was recorded on October and the minimum population (4, 3 and 0 nymphs) was observed on July. Overall population of pink, spotted and spiny bollworms was recorded as 28.80±11.90, 9.00±3.80 and 7.00±3.31 nymph, respectively. In taluka Kangarh maximum population of pink, spotted and spiny bollworms (94, 10 and 7 nymphs) was recorded on August and October and the minimum population (18, 1 and 0 nymphs) was observed on November and July. Overall population of pink, spotted and spiny bollworms was recorded as 42.20±13.85, 4.80±1.49 and 3.20±1.39 nymph, respectively. In taluka Mirpur Mathelo maximum population of pink, spotted and spiny bollworms (25, 10 and 8 nymphs) was recorded on September and October and the minimum population (1, 3 and 2 nymphs) was observed on July and November. Overall population of pink, spotted and spiny bollworms was recorded as 11.80±4.93, 6.60±2.13 and 3.00±1.54 nymph, respectively. In taluka Daharki maximum population of pink, spotted and spiny bollworms (19, 10 and 8 nymphs) was recorded on October and the minimum population (7, 3 and 0 nymphs) was observed on July.

Overall population of pink, spotted and spiny bollworms was recorded as 13.00±2.58, 4.60±1.74 and 2.40±1.46 nymph, respectively. In taluka Ubaro maximum population of pink, spotted and spiny bollworms (22, 10 and 7 nymphs) was recorded on October and the minimum population (4, 2 and 0 nymphs) was observed on August and July. Overall population of pink, spotted and spiny bollworms was recorded as 10.40±3.64, 4.46±1.43 and 2.40±1.28 nymph, respectively.

# COMPARATIVE STUDY ON THE OVIPOSITIONAL BEHAVIOR OF CAELIFERA (ORTHOPTERA) FROM THAR DESERT, SINDH, PAKISTAN

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Caelifera, low-land insects, mostly are grasshoppers. Many earning crops such as cotton, wheat, rice, sugarcane, maize and millets are damaged by them. At present, about 67 egg-pods of 04-pest species were obtained from the field. The egg-pods were identified as *Schistocerca gregaria*, *Acrotylus humbertianus*, *Poekilocerus pictus* and *Acrida exaltata*. The egg-pods have variations in size, shape and arrangement of eggs. Eggs seem sub-cylindrical and elongated in shape. Egg-pods were full of eggs without any empty spaces. Length of egg-pods of *Schistocerca gregaria*, *Acrotylus humbertianus*, *Poekilocerus pictus* and *Acrida exaltata*was calculated 45.1+2.47, 44.2+2.47, 38.2+3.07 and 43.2 +2.93 mm respectively. During this study, evidence of various species relationships through similarities and differentiation in the egg patterns has been studied with some unique result. This study might be helpful to develop a control strategy combining biotic and abiotic factors to predict when mechanical destruction of egg-pods farmers should be undertaken to prevent crop damage.

# ON THE MORPHOLOGY OF IMMATURE STAGES OF *OCHRILIDIA*, (GOMPHOCERINAE: ACRIDIDAE: ORTHOPTERA) SINDH

#### Sajjad Ali Larik, Riffat Sultana and Imran Khatri\*

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During present study different survey were carried out and 265 specimens of genus *Ochrilidia* Stal, 1873 were collected from various vegetative localities of Sindh. In order to identify various immature stages collected material has been brought to Entomological and Bio-Control Research Laboratory, Department of Zoology, University of Sindh, Jamshoro. Identification has been done through stereoscopic dissecting binocular microscope. Collected specimens of genus *Ochrilidia* were sorted out into two species viz: *Ochrilidia geniculata* (Boliver 1913), *Ochrilidia gracilis gracilis* (Krauss 1902). These species seem to be very common throughout the various agricultural fields. Huge number of various hoppers stages of these species were collected and sorted out into 05 nymphal stages. Identification keys, drawing lines and measurements of body parts were presented. This research strongly suggests that there is immense need of survey during various seasons from different habitats in order to observe the more morphological differentiations in immature stages of *Ochrilidia*.

# DIVERSITY AND SEASONAL VARIATIONS OF CAELIFERA (ORTHOPTERA) FROM KHAIRPUR MIR'S, SINDH

#### Samiullah Soomro and Riffat Sultana

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The study was planned to estimate diversity, distribution and seasonal variations of grasshoppers in croplands of Khairpur Mir's, Sindh, Pakistan. The seasonal variations were assessed by sampling 08 randomly selected sites

fortnightly during 2018-2019. The specimens were collected by hand picking and using sweep net. Adult grasshoppers were sampled from each site between 10:00 am - 01:00 PM under sunny conditions. The collected specimens were killed, identified and preserved by standard method. Data was subjected to statistical analysis and interpretations which indicate the presence of 02 families, 05 subfamilies, 06 Genera and 06 species. Family acrididae distributed in 04 subfamilies: Acridinae, Conocephalinae, Oxyinae and Cyrtacanthacridinae with 04 genera and species i-e *Acrida exaltata*, *Hieroglyphus oryzivorus*, *Oxya hyla hyla* and *Schistocerca gregaria* respectively whereas family Pyrgomorphidae offered 01 subfamily: Pyrgomorphinae with 02 genera and 2 species i-e *Atractomorpha acutipennis*, *Poekilocerus pictus*. It was noticed that *Schistocerca gregaria* and *Poekilocerus pictus* were the most abundant species with 70% whereas *Acrida exaltata* was the least common species with 3%. The study suggests that sampled area has well established grasshopper populations which require well-knitted pest management strategies to avert economic losses and conserve biodiversity. This research financially supported by Higher Education Commission, Islamabad, Pakistan (Project No. 6737 SINDH/NRPU/R&D.

# CONTROL OF *POEKILOCERUS PICTUS* (PYRGOMORPHIDAE: ORTHOPTERA) THROUGH *BEAUVERIA BASSIANA*

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At present about 225 specimens of *Poekilocerus pictus* (Fabricius, 1775) collected from the field which was found on the Akk plant. Akk Grasshopper is main agriculture pest of different crops, mainly damaging the different crops in field which are present in mostly in everywhere, rice sugarcane, wheat, maize as well as fodder particular it damage Akk plants which has medicinal valued. *Beauveria bassiana* were applied against different stages of insects. It has been observed that probably 36 hours the specimens were unable to move from one place to another with reduced feeding capability. Along this death ratio of specimens of *P. pictus* has been calculated. Among pathogens fungi have an important role in the field of bio-control agent because it has no any effect in their surrounding environment but regulating the grasshopper populations in fields. So this biocontrol agent should be applied against pest species.

# COPULATION EFFECTS ON THE OVIPOSITION AND NUMBER OF EGG PODS IN ACRIDINAE SPECIES (ORTHOPTERA: ACRIDIDAE)

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Copulation factor increase the population and grasshoppers are widely distributed having the little influence on the oviposition process and number of eggs inside egg-pods. Males are more active than females and mature earlier. Different copulatory behaviors effect on length of oviposition. Rival males sometimes show Forcible copulation even with unrelated species. Oviposition usually occurred in day time or early morning time. Before oviposition females find out the moist soil by digging. Female egg pod may also contain 50-80 eggs. 100% of offspring require 8hrs copulation without any disturbance. Egg pod may also contain 5 to 9 eggs due to unfertilized spermatophore. Sometimes only deposit foamy mass without eggs. Radially arranged eggs inside egg pods with shiny yellow color, after 3 months their color remained same in Acridinae species i.e. *Acrida exaltata* and *Truxalis fitzgeraldi* and *T.eximia exmia* 

# COMPARATIVE STUDY ON TWO SUB-FAMILIES OF EMPUSIDAE (INSECTA: MANTODEA FROM (SINDH)

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Mantids are one of the most important group of insect's fauna belonging to order Mantodea. Family Empusidae are considerable economic important, because praying mantids predatory nature, which used as biological control agent against to pest crop. Family empusidae comprised on two sub-familes Blepharodinae and Empusinae. Empusidae is second most dominant family after the family Mantidae. In extensive survey a total 85 specimens were collected from various localities of Sindh during the year 2019. All specimen belongs to two sub-families Blepharodinae and Empusinae, 38 specimens of Blepharodinae and 47 specimens of Empusidnae. Furthermore richness of species under way.

# BIODIVERSITY OF THE PYGMY GRASSHOPPERS (ORTHOPTERA: TETRIGIDAE) FROM SINDH

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Pygmy grasshoppers are the smallest representatives of the family Tetrigidae, their size is small less than 20 mm and mainly characterized by having highly elongated, tapered pronotum, that extends all over the length of abdomen. Tagmina small and scale like and may be exposed or covered by pronotum. Members of the family Tetrigidae (pygmy grasshoppers) have different body patterns usually present on the dorsal surface of the body, on their legs and also vary in colors i.e. brown grey, rusty grey or moss green in color and can be related to true grasshoppers. Pygmy grasshoppers usually found near water bodies, such as ponds, streams, pools and lakes. Occasionally they also found in dry habitats, woodlands, rice fields, mango and lemon orchards and in sandy areas with lichen. These grasshoppers especially eat roots of plants, seedlings, mosses, fungi, algae and cause considerable damage to valuable crops. During the present study 04 species have been reported, pertaining to 02 genera i.e. *Euparatettix*, Hancock, 1904, *Ergatettix*, Kirby, 1914, species belong to these genera are *Euparatettix indicus* (Bolivar, 1887), *E.sagittata* (Bolivar, 1887), *E. tricarinatus* (Bolívar, 1887) and *Ergatettix dorsifera* (Walker, 1871). The specimens were collected from grasses, rice fields, plant roots, seedlings and from the boundary of a water pool and the stones with thick mosses growing on them, at the various places while three species of genus *Euparatettix indicus* (Bolivar, 1887), *E.sagittata* (Bolivar, 1887) and *E.tricarinatus* (Bolívar, 1887) have been reported for the first time from the Sindh.

#### EFFECT OF PHASMIDS ON SOME MEDICINAL PLANTS IN NARA DESERT

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Plants consideration of *Carausius* was carried out in the year 2017 to 2018 in order to assess the food preference of *morosus* species and in result of 7 extensive surveys 87 specimens of stick insect were collected and wondering only single species i.e. *Carausius* (*Dixippus*) *morosus* (de Sinety 1902) was reported which shows its rare status in Pakistan. During present survey in the different areas of Nara desert we found that: stick insects feeding on some plants, it was surprising that some of them Plants are very objective and commonly used in the field of local

medicine. Different parts of these plants (Citrullus colocynthis, Convolvulus arvensis, Euphorbia thymifolia, Senna italic) individually and their mixture are used for treatment of different human diseases. Such as Digestive problem, skin problem toothache, stomach/abdominal pain, diarrhea, Old age diseases like joint pain, backache and sciatic pain etc.

# INTERACTION OF PREDATORS WITH APHID (SCHIZAPIS GRAMINUM) ON DIFFERENT WHEAT VARIETIES FROM LARKANA DISTRICT

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The present study was aimed to determine the interaction of predators with aphid (*Schizapis graminum*) on different wheat varieties at Larkana District from 1<sup>st</sup> January to 15<sup>th</sup> April 2018. Five wheat varieties were sown included TD-1, NIAamber, TJ-83, QS-4 and Benazir. It was observed that maximum mean aphid population was recorded on TD-1 (10.4 /leaf) and minimum on NIAamber (2.3 /leaf) in the mid of February. The maximum mean aphid population was 15.56 per ear on TD-1 in mid-March and minimum mean aphid population was 4.5 per ear on NIAamber in April. Among predators, the maximum mean population of seven spotted beetles (*Coccinella septempunctata*) was recorded on TD-1 and Benazir (1.10 /plant) and minimum population was recorded on NIAamber (0.06 /plant). The predatory species are beneficial to farmers and gardeners, as aphids destroy crops, and syrphid maggots are often used in control. The maximum population of predator Syrphid flies (Syritta pipiens) was recorded on wheat variety QS-4 (0.08 /plant) and minimum population was observed (0.05 /plant) on TJ-83 while maximum soldier beetle population (0.12 /plant) was on TD-1 and lowest population was (0.06 /plant) on NIAamber and TJ-83. This study aimed to investigate aphid population and their interaction with predators and also focused on the varietal response of wheat against aphid and their biological control agents.

# THE UNEXPECTED SCHIZOCOMICUS (SCHIZODACTYLIDAE: ENSIFERA) FROM CHOLISTAN DESERT PUNJAB

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Schizodactylidae is one unique inhabitant of desert hardly known outside entomology circles. There is only limited knowledge of the ethology and biology of these difficult-to-study nocturnal insects that still have entomologists groping in the dark. Surprisingly during the Ramadan night walk about 04 samples of Schizocomicus were collected from CUVAS University campus at 3.20 AM during May 2019. This genus first discovered on moderate slopes or level in deep layers of loose and fine sand of Nangar Parkar Sindh. It is closest member of Schizodactylidae commonly known dune cricket, which is relict group of primitive Ensifera. The specimens in hands are large in size (37-44 mm) compare to a true cricket which usually ranges from 10-25 or 30 mm. it was noticed that big mandibles and long, robust legs with expanded tarsi give it a somewhat menacing appearance. I examined that its paddle-like projections is not only an adaption to aid its burrowing nature but also serves to support its large body as it hunts for prey in the sand. As this genus was spotted near the Cholistan desert, where there is no river or water canal nearby. But why insect choose a punishing desert for home? No one knows yet. How do these crickets survive in the harsh desert conditions? in my opinion usually Dune crickets (both adults & nymphs) dig isolated tunnels each one for itself - in the sand, where they stay all day protect themselves from not only natural enemies but this tunnel make proper supply of water taken from fog, dew and rainfall. Tunnels made by dune crickets are designed to let water droplets flow inside however, detail study on this parameter is underway. But collection of these samples from Cholistan desert extend its distribution range from Thar-Nara to Cholistan but also constructed new record for Punjab. The present study recommends that incident of Schizocomicus is a testimony to the fact that the diminutivelooking dune crickets are a force to reckon with in the desert ecosystem – armed to protect and sustain themselves in climes even humans struggle to survive. Financial assistance received form Higher Education Commission (HEC) Islamabad, Pakistan under Project No. 6737 SINDH /NRPU /R&D/.

# EFFECT OF CHLORPYRIFOS AND ACEPHATE ON WEB BUILDING BEHAVIOR OF ORB WEB-SPIDER (Araneae; Araneidae)

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Traditionally, different agricultural practices are being used to enhance agricultural yield along with insecticides. Excessive use of insecticides in crops has deleterious effect on natural predators, such as spiders. They reduce their efficiency as biological control agents. However, estimated harmful effects of insecticides Chlorpyrifos and Acephate on the web building behavior of orb-web spider have been checked with different time gap. Both insecticides effect spiders and their web building and Chlorpyrifos proves lethal for spider survival as it causes 70 percent mortality at recommended field rate concentration while Acephate causes only 50 percent mortality. Finally, to select those insecticides and insecticidal concentrations that have least effect on natural behavior of spiders to construct their webs so that these environment friendly natural enemies of pests can be conserved and also can be enabled to continue playing their role in controlling pest population.

### TOXIC IMPACT OF IMIDACLOPRID AND LEAD ON DETOXIFYING ENZYMES OF SILKWORM BOMBYX MORI

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Silkworms are economically important insect and badly effected by xenobiotics like imidacloprid and heavy metal. They are reared very carefully in the indoor insectaries and mostly exposed to different chemicals through food contamination. In this study, role of detoxifying enzymes in the susceptibility of Imidacloprid (commercial formulation and technical grade) and heavy metal Lead (Pb) were evaluated in 5<sup>th</sup> instar larvae of *Bombyx mori*. For this purpose larvae were exposed to different concentrations of Imidacloprid and Pb and mortality were recorded after 24h of exposure. The LC<sub>50</sub> values of these xenobiotics were estimate and changes in quantity of different enzymes of hemolymph were assessed. The calculated acetylcholinesterase (AchE) in control group and Pb treated group was limited but high in imidacloprid commercial (formulation or technical grade). Silkworm larvae treated with mixture of xenobiotics also have reduce AchE quantity. Catalase (CAT), Glutathione-S-transferase (GST) and Superoxide dismutase (SOD) concentration was high in the mixture of Imidacloprid and Pb as compared to other treatments. The level of these enzymes in Pb treated larvae and control larvae did not differ significantly. Present study demonstrated that detoxifying enzyme such as AchE, SOD, CAT and GST were involved in the free radical scavenging produced in response to xenobiotics exposure in larvae of *B. mori*.

#### THE INFLUENCE OF HOST PLANT ON THE POPULATION GROWTH OF BACTROCERA DORSALIS

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The concentration of phytofagous and monophagous insect species feeding on variety of host plants, this thought paid the vast diversification on the pest population and consumption of host plant. The family Tephritidae are perfect for study of the influence for furgivorious consumption and in community of phytophagous insects. This

family includes all the fruit flies which are economically important because many species in family Tephritidae attack the cash crop of fruits and vegetables worldwide. The influence of conservational conditions; such as host plants and weather parameters factors on the richness and inhabitants. The study of *Bactrocera dorsalis* population were worked out in major host of mango orchards. In many districts of Sindh such as Naushahro Feroze, Khairpur Larkano, Hyderabad, Matiari, Tando Muhammad khan Mirpur Khas etc. During the experimental study *Bactrocera dorsalis* were observed in 2017-2018 from, Jan-December. It emerging time at the last week of May. Crowning incidence of fruit fly activity was observed during the mid of July. However, a gradual increase was observed during the beginning of June and slowly gradually decline during the mid of August. Correlation studies between host, and pest showed significant positive relationship. Availability of Host (mango fruits) were another crucial cause for influencing population availability.

#### ANTS OF GENUS LASIUS FABRICIUS, 1804FROM PAKISTAN

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The genus Lasius Fabricius, 1804 is bulging and familiar ant genus, it is probably best known for its conspicuous nuptial flights, its habit of tending and transporting homopterous insects, and the temporary parasitic behavior of some of its species. Lasius are timid species adapted to living under the bark and in the wood of tree trunks. Large population of this species are known to inhabit in the Forest and limited to living trees, which the ants penetrated from the trunk up into the main branches and down into the roots, trees include; oaks, elm, ash, beech, poplar and maple (Donisthorpe 1927). For present studies the material was collected by the first author during in 2016- 2017 from Pakistan. Information on elevation was obtained from Google Earth Pro v. 7.1.8.3036 based on coordinates. The main sampling method used to collect ants was hand collection targeting nests. Photograph of specimens were taken using Leica DFC450 camera mounted on a Leica M205 C dissecting microscope. Stack imaging was performed with 40 to 50 images per montage of the specimens taken, enhanced and with specimens measured using the Leica Application Suite v. 4.5. Currently, 09 species of the genus Lasius are described from the high-altitude territories of Pakistan, it may be assumed that the actual number of species found in this region will be much larger. In addition, we believe that the Himalayas are another center of diversity of this genus. The territory of Pakistan in myrmecological terms is very poorly known, the Author first time collected this genus from upper highaltitude part of Pakistan and shows distribution characters and keys of all the species of genus. The audience will have incite to the diagnosis, distribution and ecology of the genus *Lasius*.

#### CURRENT STATUS OF THRIPS OF TANDOJAM

### Manzoor Ali Khushk<sup>1</sup>, Imran Khatri<sup>1\*</sup>, Fahad Nazir Khoso<sup>1</sup> and Zubair Ahmed<sup>2</sup>

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Thrips (Thysanoptera) are slender bodied small insects range from 0.5 to 2.5 mm, with piercing and sucking mouthparts, adult thrips are attracted to flower scent and colour, whereas, larvae feed on leaves, fruits, liquids and pollen, adults do not stay on their breeding sites to feed. Thysanoptera consists of 6000 described species of thrips. Thrips being an important pest, pollinator and predator plays an important role in agriculture, but our knowledge on this insect is very weak. The first comprehensive work on thrips from Pakistan was published by Palmer (1992). Later, some baseline studies conducted by various authors on description and incidence include. Looking at the importance of thrips present studies were proposed to conduct. For present studies thrips were collected from Agricultural fields, floricultural fields, orchards, ornamental plants, horticultural gardens etc. from Tandojam and

adjoining areas. Specimens were collected by beat methods. These were put into a AGA, a mixture of 10 parts of 60% ethyl alcohol with 1 part of glycerin and 1 part of acetic acid. The maceration were performed to remove fat bodies and soft connective tissues: For habitus (adult) images the high pixel camera was used, and for the images of Habitus of thrips, camera fitted on microscopes a) HT (40X), b) Kyowa Medilux 20 were used. Microscope images were processed through software combine ZP and Helicon focus for getting whole focused image. Thrips were identified through the available keys. The audience will have incite on thrips species collected and identified from various fields of Tandojam.

#### SOIL INSECT FAUNA OF ROSE CROP

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The survey of rose fields was conducted at different locations during the year 2019 and collections of ground dwelling insects associated with rose plant was made in daytime from the garden of Faculty of Crop Protection, Sindh Agriculture University Tandojam. In present study total 106 specimens of class Insecta were collected from soils of the rose plant from Faculty of Crop Protection garden, the identification revealed the occurrence of various 09 species under 03 insect orders; Coleoptera, Linnaeus 1758, Orthoptera, Latreille, 1793 and Hymenoptera, Linnaeus, 1758. Order Coleoptera revealed the occurrence of 04 species including; *Chlaenius* sp. under family: Carabidae, Latreille, 1802 and subfamily Licininae, Bonelli 1810; family Scarabaeidae, Latreille, 1802 discovered with two species records; *Aphodius ghardimaouensis* (Balthasar, 1929) under Subfamily Aphodiinae Leach, 1815 and tribe Aphodiini Leach, 1815; *Xyloryctes* sp. under subfamily Dynastinae, MacLeay, 1819 and tribe Oryctini, Mulsant, 1842; *Anthicus crinitus* Laferte, 1849 family Anthicidae, Latreille, 1819, subfamily Anthicinae Latreille, 1819 and tribe Anthicini Latreille, 1819. Orthoptera with two species record *Gryllotalpa africana* (Palisot de Beauvois, 1805) under family Gryllotalpidae Saussure, 1870 and *Acheta domesticus* (Linnaeus, 1758) under family Gryllidae Laicharting, 1781. Hymenoptera was discovered with 3 species of family Formicidae Latreille, 1809 and subfamily Formicinae Latreille, 1809 including; *Camponotus sp., Camponotus compressus* (Fabricius, 1787) and *Paratrechina longicornis* (Latreille, 1802).

# HIGH TEMPERATURE EFFECT ON SURVIVAL OF *DYSDERCUS KOENIGII* (F.) (HEMIPTERA: PYRRHOCORIDAE)

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Under natural environmental conditions, insects are highly subjected to thermal stress. Data concerning the effects of high temperature on the survival of *Dysdercus koenigii* (F.) are scarce. In the present work, the survival rate of all immature stages and adults of *D. koenigii* exposed to very high temperatures were studied. All stages were exposed to four high temperatures (40, 42.5, 45 and 47.5°C) for time periods ranging from 1.00 to 420 minutes, depending on the temperature treatment. Insects were then returned to normal temperature conditions. The survival rate of all stages decreased with the increase in the time of exposure at all temperatures examined. The lethal time needed to cause mortality in 50% and 90% of all stages decreased rapidly with the increase in the temperature from

40 to 47.5°C. Based on these results, we concluded that the survival rates of all stages of *D. koenigii* were significantly affected when they were exposed to high temperature stress.

# EFFECTS OF TWO PREDATORY ANTS CAMPONOTUS COMPRESSUS AND MESSOR HIMALAYANUS (FORMICIDAE: HYMENOPTERA) AND THEIR PHEROMONES ON MANGO ORIENTAL FRUIT FLY BACTROCERA DORSALIS

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Mango oriental fruit fly, Bactrocera dorsalis L. (Tephritidae: Dipteran), is most damaging pest of mango in tropical and subtropical regions throughout the world. To prevent from severe losses of mango, it is necessary to manage the B. dorsalis. Chemical control gives effective and rapid control against B. dorsalis but chemical control developed resistance problems and environmental issues. Therefore, it is necessary to adopt such alternate control measures to manage the B. dorsalis that are environment friendly cost effective and have no effects on both quality and quantity of mango. Biological control is known as alternate effective control method because it is environment friendly and has no residual effect. In present study, two ant species Camponotus compressus (Fabricius) and Messor Himalayanus (Forel) used as biological agent in field and laboratory trials. Mango were exposed with different number of ants (25, 50, 75, 100, 150, 200, 300, 400 and 500). B. dorsalis flies were allowed for oviposition, number of landing, time spent and on the mango fruits. Larvae and pupae of B. dorsalis were provided as a pray in Petri dish. In Y-tube olfactometer, behaviour of B. dorsalis against exposed and unexposed mango was recorded. Results showed that female adult ovipoisted five times more on unexposed mango as compared to exposed mangoes. Ants pheromones change the behaviour of B. dorsalis and also affect the number of landing and time spent on the fruits. Significant predation was observed on the larval and pupal stage of B. dorsalis by both ants species. Ants pheromones also effect the olfactory response of B. dorsalis at adult stage. So, ants might be use as a good predator for B. dorsalis and their pheromones use as repellent in future to control B. dorsalis. Need more extensive study to detect the exact compound responsible for repletion.

#### EVALUATION OF DIFFERENT X-RAY DOSES ON DEVELOPMENT OF FRUIT FLIES, BACTROCERA SPP. FOR INSECT STERILIZATION

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Fruit flies are the most destructive pests of the fruits grown throughout the world. Among various species of fruit flies, *Bactrocera zonata* and *Bactrocera dorsalis* are the predominant species attacking peach, guava, mango and other fleshy fruits in Pakistan. Their control largely depends on the application of insecticides, which pose potential health risk in gardens, destruction of beneficial insect fauna and development of insect pest resistance. Research on fruit flies is therefore, geared towards development of alternative control strategies. In the present studies the strategies were made to determine the effect of different X-ray doses such as 20, 40, 60, 80 and 100 Gy on the pupae of the *B. zonata* and *B. dorsalis*. The tests were conducted on laboratory strains of the two *Bactrocera* species reared on mango, guava and artificial diet. After x-ray irradiation treatment, data were taken on partial pupal emergence, deformed emergence (Male and Female), Normal emergence (Male and Female) and unemerged pupae of both species. Peach fruit fly was the most tolerant of the both species to irradiation. Whereas, laboratory strains of

each species when reared on natural diet were equally tolerant of irradiation, as compared with artificial diet. An irradiation dose of 100 Gy resulted in most number of unemerged pupae. Results of the present investigation would be useful in developing suitability of X-ray irradiators to integrate the SIT programme in the orchard agro-ecosystem.

# ECOLOGY, DIVERSITY AND DISTRIBUTION OF SPIDERS FAUNA (ARACHNIDA: ARANEAE) OF DISTRICT MOHMAND, KHYBER PAKHTUNKHWA, PAKISTAN

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The study was conducted during 2018-2019 in district Mohmand, Khyber Pakhtunkhwa, Pakistan. During the present study, spiders specimens were collected from three sites Wheat crops, Maize crops, refuges of mountain. The pitfall traps, hand collection and vacuum succession were used for sampling. A total of 1231 specimens were collected the present study aimed to improve the understanding of species distribution, diversity and seasonal variation in Mohmand. Diversity Indices were calculated including indices for species richness, species evenness, and Shannon – Weiner Diversity index (H') and Simpson's Index . The more diversity of spiders communities were observed in refuges of small mountains. Moreovere, populations of immature and adults were more dominant in maize and Wheat crops at different intervals of the season. Threats to shifting of biodiversity were observed land conversion, desertification and anthropogenic activities were observed the major threads for degradation of taxa diversity at district Mohmand.

### DIVERSITY AND DISTRIBUTION PATTERN OF FOLIAGE ARTHROPODS IN FRAGMENTED ARABLE LANDSCAPES

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This study was designed to explore the "diversity and distribution pattern of foliage arthropods in fragmented arable landscape. For the assessment of arthropods diversity, two fragmented arable patches were selected, and samples were collected by sweep net, forceps and direct hand picking, then sorted and labelled accordingly. Identification was done up to specie level based on morphological characters. Total 3376 specimens were collected, from which vegetable landscape comprised of 2256 specimens with 8 orders, 49 families, 80 genus and 90 species and from Nursery landscape, total 1120 specimens were collected comprising of 8 orders, 33 families, 52 genus and 60 species. Maximum relative abundance 7.41% (N = 83) was recorded from Nursery landscape for *Harmonia octomaculata* species while, from vegetable landscape highest 2.17% (N = 49) was recorded from the vegetable landscape (4.4998), (0.0207), (0.9792), (45.494) respectively as compared to Nursery. ANOVA results among both landscapes showed significant results. Regression analysis showed that the diversity of arthropods varied significantly in both arable landscapes.

#### DIPTERAN POPULATION IN DOMESTIC AND NATURAL TERRITORY

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Diptera (Insecta) is one of the largest order having 150,000 different types of flies are now known and estimates are that there may be more than 1,000,000 species living today. They are found in all terrestrial habitats in the world apart from Antarctica. They play a key role in pollinating flowers. In particular, mosquitoes represent a key threat for millions of people worldwide. The purpose of research work was to illustrate the dipteran population in different territories (Domestic territory from Rahim yar khan and Natural territory from Fatima Hall University of Agriculture Faisalabad). Triplicate water tubs containing 10% formalin solution were placed under the light spot were used to collect the nocturnal insects from dawn to dusk. White light traps were set at distance of 06 ft from each other. Collection was done by direct hand picking and with the help of forceps. Specimens were preserved in 70:30% alcohol and glycerin solution. The collected samples were identified and sorted with the aid of naked eye, magnifying glass and light microscope. All the specimens were identified up to the species level. After completing the whole research, total 227 specimens were noted and maximum population was recorded from Domestic territory 54.63% pertaining to 1 orders, 12 families, 14 genera and 14 species; Wherein least population was recorded from Natural territory 45.37% pertaining to 1 order, 8 families, 9 genera and 9 species. The overall data from domestic territory, Musca domestica was recorded as an unusual contributing species and followed by Hermetia illucens, Fannia spp., Hermetia spp., Milichiidae spp., Cuterebrinae spp. and Simulium trifasciatum was also recorded. From Natural, Egle spp. was recorded as highly abundant species and followed by Hebecnema umbratica, Anopheles stephensi, Beris geniculate, Cordilora ciliate, Leptocera spp., Haematopota pluvialis, Culiseta longiareolata and Glossina spp.

#### POPULATION STRUCTURE OF ORDER DIPTERA IN MIXED-CROP ZONE

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Diptera is one of the major insect orders containing 1,000,000 species including horseflies, crane flies and hoverflies, have a significant effect on agroecosystems. Among them, some leaf-miner flies, fruit flies and gall midges are considered as major pests of agricultural crops. Relying upon their diversity and significance in different crops zone, research was conducted for three months (July 2017 to September 2017) in Faisalabad, Punjab, Pakistan. Dipteran fauna was collected from an area of 3 hectare for two hours during 07:00am to 09:00am with the help of sweep net, direct handpicking and forceps from mixed crops weekly. Collected specimens were preserved in containing 70: 30% alcohols and glycerin solution and then shifted to the Biodiversity laboratory, Department of Zoology, Wildlife and Fisheries, University of Agriculture Faisalabad for further systematic studies. The collected samples were identified and sorted with the help of: Naked eye, Magnifying glass and Light microscope up to the specie level. Total 139 specimens were collected belonging to 1 order, 22 families, 39 genera and 45 species. Among them, Musca domestica (Muscidae: Diptera) was recorded as a highly abundant species with relative abundance, followed by Periscepsia carbonaria (Tachinidae: Diptera), Aedes albopictus (Culicidae: Diptera) and Blepharotes splendidissimus (Asilidae: Diptera), Drosophila pseudoobscura (Drosophilidae: Diptera) and Calliphora vicina (Calliphoridae: Diptera). The genus Musca and Drosophila were recorded most abundant followed by Calliphora, Periscepsia, Aedes and Blepharotes, Simulium. As well family Muscidae, were recorded as highly abundant followed by Calliphoridae, Drosophilidae and Tachinidae.

# CHRONOLOGICAL ALLIANCES OF FOLIAGE INSECTS FROM SUMMER FLOWERS IN DOMESTIC AND WILD TERRITORY

### Khadija Hameed, Naureen Rana\*, Hafiza Aqsa, Nazia Ehsan, Saher Shabbir, Imran Ahmad Raja, Sobia Kanwal and Shahla Nargis

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Biodiversity is the variety of all forms of life, its more prominent part is insects as they are world's most diverse group of animals represent 58-67% of known global biodiversity. Flowers represent high quality resources to many insects and some species benefit the host plant by transporting pollen and may utilize flowers for a variety of reasons. This study was conducted to find out the Chronological alliances of foliage insects from summer flowers in domestic and wild territories of Faisalabad, and the research sites were: D-ground University of Agriculture, Faisalabad and Fatima Hall, University of Agriculture, Faisalabad. Sampling was done once in a week by selected the area of One acre with the help of sweep net, forceps and direct hand picking method, collection were made for two hours during 07:00 am to 09:00 am from both territories after that specimens preserved in stock jars containing 70: 30% alcohols and glycerin solution and then shifted to biodiversity laboratory, Department of Zoology, Wildlife and Fisheries, University of Agriculture Faisalabad for further systematic studies. Collected specimens were identified and sorted on the basis of their morphological characters with the help of naked eye, magnifying glass and light microscope, identification was done up to specie level according to taxonomic material. The results showed total 64 specimens were collected from wild belonging to 10 orders, 15 families, 18 genera and 20 species, while 82 specimens were collected from domestic belonging to 9 orders, 16 families, 20 genera and 20 species. From the overall data, Camponotus Empedocles and Zizula hylax were recorded as highly abundant species with 15.63% (N=10), followed by Polistes olivaceus and P. carolina 14.06% (N=9) from D-ground while from Fatima hall, Coreus marginatus was recorded highly abundant with 24.39% (N= 20), followed by Annelid spp. 14.65% (N= 12), Zizula hyalyx and Musca domestica. From D-ground relative abundance upto family was concerned in case of D-ground Vespidae 28.13% (N= 18), followed by Formicidae 25.00% (N= 16) and from Fatima hall family Coreidae 24.39% (N = 20), followed by family Formacidae 20.73% (N = 17) and Nereidae 14.63% (N = 12).

### ECOLOGICAL DISTRIBUTION OF NOCTURNAL INSECTS AT THE WET TERRITORY

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Insects are the key elements of the ecosystem that plays an important role in sustainability of ecological balance. This research was organized to find out Spatio-Temporal diversity of nocturnal insect population and communities at different wetlands of fish farms by considering their ecological patterns and conditions in under the ecological condition of Ali Pur Chatha (APC) and Faisalabad (FSD). Maximum population was recorded from wet territory of APC as 83.82% (N= 14862) and least population was recorded from Wet territory of FSD within UAF 16.17% (N= 2867). 11 orders, 68 families, 123 genera and 140 species had been recorded from wetland of APC while 10 orders, 77 families, 145 genera and 177 species had been recorded from wet territory of FSD. At genus level for *Enochrus* 25.39% (N = 3773) from APC while from the wet territory of Faisalabad for the genera *Tanytarsus* 34.01% (N = 975). From total 11 orders collected from wetland of Ali Pur Chatha, highest Relative Abundance was resulted out for Coleoptera 43.32 percent (N= 6438) while from ten orders being collected from wet land of Faisalabad, maximum Relative Abundance had been calculated for Diptera 61.39% (N = 1760). (H') had been resulted out maximum (3.60001) from wetland of Faisalabad and minimum from Ali Pur Chatha (2.74730). (H<sub>max</sub>) had been documented maximum from Faisalabad (5.1761), Evenness was recorded highest from wet territory of Faisalabad (0.6955) and Dominance was seen maximum from wet territory of Ali Pur Chatha (0.4440). However, (R) was documented highest in wet land of Faisalabad (88.9971).

# ACCEPTABILITY OF NOCTURNAL INSECTS REGARDING DIFFERENT SOLUTION

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Insects are the largest and most diverse group of hexapod invertebrates within the arthropod phylum; having more than two million described species and represent more than half of all known living organisms on earth biosphere. They are the pollinators, undertakers, leaf litter sweepers, garbage collectors, soil conditioners and natural fertilizer producers. They found in all ecosystems with different classification e.g. order Coleoptera, Diptera, Hymenoptera and Lepidoptera etc. Nocturnal insects become active at night and sleep or less active during the daytime. The research was conducted to record the Acceptability of nocturnal insects regarding different solutions under the territory of district Faisalabad, three light traps with different solution e.g. sugar solution, 10% formalin solution and distal water were used. Light traps were set at distance of 6 ft. apart from each other. Collection was done by direct hand picking and with the help of forceps. Specimens were preserved in 70:30% alcohol and glycerin solution. The collected samples were identified and sorted with the help of naked eye, magnifying glass and light microscope. All the specimens were identified up to the species level. After completing the whole research trials as per methodology, in case of formalin solution, 210 specimens were collected belonging to 8 orders, 16 families, 20 genera and 21 species while in case of sugar solution, total 328 specimens were collected belonging to 8 order, 18 families, 22 genera and 19 species and in distal water solution, total 213 specimens were collected belonging to 8 order, 15 families, 17 genera and 19 species. From formaline solution, Apis nearctica was recorded as an extraordinary contributing species with relative abundance of 40.95%, followed by Phyllophaga marginalis 20.95%, Anomalon spp. 9.52% and Tineola bisselliella. From sugar solution, Apis nearctica was recorded as an extraordinary contributing species with relative abundance of 33.2% followed by Apis cerana 11.28%, Tortrix viridana 10.06% and Anomalon spp. 9.45%. From distilled water solution Anomalon spp. was recorded as an extraordinary contributing species with relative abundance of 30.05% followed by Apis cerana 15.49% and Condica illecta 13.62%. The most diversity found in sugar solution followed by distilled water solution and formaline solution respectively.

# EFFECT OF MATING INTERLUDES AND MINI NUCS SPACING OF HONEY BEE (APIS MELLIFERA L.) ON THE DRIFTING OF QUEENS RETURNING FROM NUPTIAL FLIGHTS

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Queen rearing is hardworking and time taking process. The loss of honeybee queens during nuptial flights upsurges the cost of their production. It was hypothised that to examine if the spacing of nucs influences the drifting of queen honey bees, which return from mating flights and also compared the drifting of in two mating interludes March and April 2019 *Apis mellifera* L. queens. Total of 63 queens were studied which were placed in mini mating nucs together with about 300-350 Nurse worker bees collected from the unsealed brood frames. Some of the mating nucs were arranged in rows spaced 30 cm apart, without any landmarks, and other nucs were spaced a few meters apart, next to trees or bushes randomly. Each group of mating nucs was tested in March and April. The results show that significantly more queens failed to return from mating flights to nucs placed in rows without any landmarks (49%) than from those placed randomly next to trees or bushes (9%). The study also showed that there is no significant differences between level of drifting during various raring interludes. It was concluded from the results that mainly more queen bees are lost during nuptial flights from mating nucs arranged in rows without landmarks compared to nucs placed next to trees or bushes. The mating interludes does not affect the drifting of queens returning from mating flights.

# EFFECTS OF ORGANIC ARTIFICIAL FERTILIZERS ON FITNESS COST OF CULEX QUINQUEFASCIATUS SAY

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Culex quinquefasciatus Say is a cosmopolitan distributed pest. It is a major vector of diseases in Asia. Experiment was conducted in the laboratory to assess the effects of artificial nitrogenous fertilizers *i.e* Urea, Nitrophos and Diammonium phosphate on the various fitness parameters of Cx. quinquefasciatus. Fifty 1<sup>st</sup> instar larvae were reared in different media of fertilizers, four different strains i.e., urea strain, NP strain, DAP strain and field strain were named. Larvae were monitored throughout their immature stages. Larval duration, pupal duration, pupal weight, survival from 1<sup>st</sup> instar to pupae and emergence (%) were recorded. The survival of immature from 1<sup>st</sup> instar to pupae was lower in urea and NP strain then Field and DAP strain. Developmental time was reduced in DAP strain. None of the all fertilizers dose had significant impact on emergence of immature of Cx. quinquefasciatus. These findings demonstrate that urea and NP fertilizers are toxic for the immature of Cx. quinquefasciatus.

#### URBAN PARKS AS HOTSPOTS FOR SUPPORTING BIODIVERSITY OF URBAN POLLINATORS

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Urban and suburban landscapes sites can provide refuges for biodiversity of bees and other pollinators. Urban landscapes have to low biodiversity as compared to peri-urban sites, but a few studies recommend that few urban land uses can support the considerable pollinator populations. There is immense interest in planting urban settings to benefit pollinators, with the potential that gardens and urban areas could act as wide system of pollinator-friendly habitats. However, there are great plant cultivars accessible to the gardener. The abundance and diversity of pollinators ensures the pollination and ecosystem services and diversity. As previous studies have shown that bee abundance and diversity is subjective by the composition and the structure of the surrounding landscape and quantified bee visitation to 72 species of flowering landscape woody plants across 373 urban and suburban sites in USA, sampling and identifying the bee assemblages associated with 45 of the most bee-attractive species. In another study, insect visitation 111 different ornamental plant cultivars to meter square plots at a site in central UK that contain over 9000 pollinator observations, which were identified to species as 'solitary bees', Syrphidae, Lepidoptera and 'others'. It is also found that there was no difference in numbers of insects attracted to native or non-native plant species, or according to whether plants were annuals, biennials or perennials, but found that native plants involved in a significantly higher diversity of flower-visiting pollinator insects. Public awareness of waning pollinator has improved interest in growing plants that provide floral resources for bees. Many publications and websites list "bee friendly" plants, but such lists are not usually based on empirical data, nor do they emphasize flowering trees and shrubs, which are a major component of urban landscapes. These data would help city foresters, landscape managers, and the public make informed decisions to create bee-friendly urban and suburban landscapes.

#### KATYDIDS (TETTIGONIIDAE: ORTHOPTERA) OF PAKISTAN

### Waheed Ali Panhwar<sup>1\*</sup>, Riffat Sultana<sup>2</sup> and Muhammad Saeed Wagan<sup>2</sup>

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As a result of this work 47 species of Tettigonioidea pertaining to 07 sub-families i-e: Pseudophyllinae, Phaneropterinae, Conocephalinae, Tettigoniinae, Hexacentrinae, Mecopodinae and Decticinae belonging to 20 tribes and 22 genera have been studied from Pakistan. The species Sathrophyllia nr. rugosa (Linnaeus, 1758), S. femorata to sub-family Pseudophyllinae, surprisingly, sub-family Phaneropterinae possess maximum new records ie: Trigonocorypha unicolor, Stål, 1873 T. angustata, Uvarov, 1922 ,T. nr. angustata, Phaneroptera bivittata B-Bienko, 1954, .Holochlora nigrotympana Ingrisch, 1990, H. venosa Stål, 1873, H. astylata Karny, 1926, Ducetia japonica, Thunberg, 1815, Letana rufonotata (Serville, 1838), L. bulbosa Ingrisch, 1990, Tylopsis lilifolia Fabricius, 1793, Isopsera spinosa Ingrisch, 1990, .I. stylata Brunner von Wattenwyl, 1878, I. pedunculata Brunner von Wattenwyl, 1878, I. astylata Karny, 1926, Himertula kinneari (Uvarov, 1923) and H. marmorata (Brunner von Wattenwyl, 1891) and there are 4 new records in sub-family Tettigoniinae viz: Calopterusa balucha (Uvarov, 1932), Platycleis grisea (Fabricius, 1781), Glyphonotus sinensis Uvarov, 1939 and Eupholidoptera karatolosi Mofidi & Quicke 2007 and 2 each to sub-family Hexacentrinae, Mecopodinae and Decticinae, Hexacentrus unicolor Serville, 1831, H. pusillus Redtenbacher, 1891, Mecopda platyphoea Walker, 1870, Afromecopoda monroviana (Karsch, 1886), Decticus verrucivorous (Linnaeus, 1758) and D. Albifrons (Fabricius, 1775). During the present study it was also observed that Phaneroptera spinosa, Bei-Bienko, P. roseata, Walker of Phaneropterinae and Conocephalus maculates (Le-Guillou) and Euconocephalus incertus Walker to Conocephalinae were widely distributed species, the possible reason of this spread might be feeding on nutritional rich vegetation of favourable climatic condition of region. Besides this, single individual each of *Phaneroptera gracilis* Bei-Bienko, Trigonocorypha nr. angustata and Letana rufonotata (Serville) of Phaneropterinae: Platycleis intermedia (Serville), Tettigonia caudata (Charpentier), Glyphonotus sinensis Uvarov, Eupholidoptera karatolosi Mofidi & Quicke to Tettigoniinae were considered as rare. In addition to this, Tylopsis lilifolia Fabricius was restricted to Baluchistan. Further, information on the distribution and ecology of the all species is given and a key to Pakistan species of Tettigonioidea is presented. Moreover, the record collection of sub-families Phaneropterinae and Conocephalinae represents the first comprehensive record of the entomological fauna of Pakistan. It was noted that the distribution of all previously recorded species has been greatly extended to the localities. The taxonomic keys for various taxa have also been constructed for their future identification. Nevertheless, a brief description of each supra-generic category of Tettigonioidea along with photographs and synonymy is also documented. Additionally, detailed list of host plants from Pakistan was also composed for the first time.

### STUDY ON BIOACOUSTICS OF GRYLLIDAE (ORTHOPTERA) OF SINDH PAKISTAN

#### Insaf Ali Veesar and Waheed Ali Panhwar\*

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The Gryllidae is an important member of Ensifera. The Grylloids bears somewhat similarities with Tettigonioidea but it has 04 segmented tarsal in female, while ovipositor is needle like of in cylindrical shaped, in appearance straight of curved it comprised on 02 pairs of valves. Their cerci elongated, flexible while few species are greenish in color. They are nocturnal they hide themselves during extreme weather and live in humid environment during day period. Bioacoustics is modern techniques used to identify the insects on the basis of their sound productions for the purpose of mate call or food call. Crickets belongs to family Gryllidae are pest of agricultural lands. They produce sound for the different purpose such as food call, mate call or to show their existence in the particularly locality etc. During the present study, sound of various species of crickets were recorded with help of Linear PCM recorder on 96 kHz 24bits. We analyzed the recorded data on the software that show different wavelength and different curves. Some of the species produces high frequency sound in various ranges and some

species produces low frequency sounds. Present bioacoustics mechanical technique is for the first used in insect identification in Sindh Pakistan.

# BIODIVERSITY OF PYGMY GRASSHOPPERS OF DISTRICT NAUSHAHRO FEROZE, SINDH

#### Mehar-un-Nisa Maitlo and Waheed Ali Panhwar\*

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Pygmy grasshoppers are generally more diverse groups among Orthopterans due to their small size and unique appearance that make them more remarkable among other Orthopterans in morphological point of view. These little creatures are mostly ground dwelling and generally come across among leaf-litter in the floor of forests or in moist places, swampy habitats neighboring water ways, rivers or up ended water. Pygmy grasshoppers were collected from different talukas (agricultural fields and their surrounding vegetation) of district Naushahro Feroze through traditional insect net as well as by hand picking. A total of 186 specimens which were collected during the October 2018- July-2019. The specimens were identified into Family Tetrigidae, two subfamilies: Tetriginae and Scelimeninae with 05 genera and 07 species: *Hedotettix gracilis* (Haan, 1843), *Hedotettix lineifera* (Walker, 1871), *Hedotettix punctatus* Hancock, 1909, *Paratettix meridionalis* (Rambur, 1838, *Ergatettix dorsiferus* (Walker, 1871), *Thoradonta spiculoba* Hancock, 1912 and *Scelimena razalii* Mahmood, Idris & Salmah, 2007. Beside this distribution of species along with habitus images and description is provided. Hopefully, this study will contribute a brick to the knowledge of pygmy grasshoppers from this region.

# MORPHOLOGICAL AND DISTRIBUTIONAL NOTES ON ACRIDIDAE (ORTHOPTERA) FROM DISTRICT DADU, SINDH.

### Preh Razaque Memon and Waheed Ali Panhwar\*

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Acrididae are commonly known as true grasshoppers. They are diverse group amongst order Orthoptera . The grasshoppers belonging to family Acrididae are pests of agricultural crops, forests, vegetables, orchards and wide variety of fruits. The true grasshoppers were captured from four talukas of district Dadu i-e: Taluka Dadu, Taluka K.N.Shah, Taluka Mehar and Taluka Johi during October 2018- July 2019. The collected specimens were 213 which were sorted out into Family Acrididae and 04 subfamilies i-e: Acridinae, Oxyinae, Oedipodinae and Cyrtacanthacridinae falling into 06 genera and 06 species i-e: Oxya velox (Fabricius, 1787), Cyrtacanthacris tartarica (Linnaeus, 1758), Anacridium rubrispinum Bey-Bienko, 1948, Acrida exaltata (Walker, 1859), Truxalis eximia eximia Eichwald, 1830 and Aiolopus thalassinus thalassinus (Fabricius, 1781). The highest population was observed of Oxya velox 27.23% followed by Acrida exaltata with 22.53% and Anacridium rubrispinum with 15.49%. While lowest population was observed in Truxalis eximia eximia with 9.38% followed by Cyrtacanthacris tatarica with 12.20% and Aiolopus thalassinus thalassinus with 13.14%. Beside this, morphological description along with distributional data and photographs are provided.

### ON THE MORPHOLOGY OF FIELD CRICKETS (GRYLLINAE) FROM DISTRICT KHAIRPUR

### Qurat-ul-Ain Dharejo\*, Abdul Manan Shaikh and Waheed Ali Panhwar

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The field crickets belongs to subfamily Gryllinae family Gryllidae and order Orthoptera. They vary in their size from 15 to 25mm, usually with black or brown in their body coloration. Both the male and female may have similar

size. Female species can be distinguished morphologically by the presence of sword or spike shaped ovipositor. Males can distinguished by the presence of cerci present at the end of abdominal segments. The females use their ovipositor to deposit their eggs in the grounds in order to protect from predators and preys Field crickets were collected from agricultural fields and their surrounding vegetation of district Khairpur Mirs through traditional insect net as well as by hand picking. The collected specimens were 167 which were sorted out into single sub-family Gryllinae falling into 05 genera and 07 species i-e: *Gryllodes sigillatus* (Walker, 1869), *Gryllodes supplicans* (Walker, 1859), *Gryllopsis histrio* (Saussure, 1773), *Acheta burdigalensis* (Latreille, 1804), *Acheta domesticus* (Linnaeus, 1758) with 36% and *Gryllodes supplicans* (Walker, 1859) with 24%. While minimum population of *Gryllopsis histrio* (Saussure, 1877) with 3% and *Acheta burdigalensis* (Latreille, 1804) with 5% respectively. Additionally, detail description of species month wise distribution is provided. Finding of field crickets from this area will be instrumental for the farmers and researchers concerned with the biodiversity of this group.

# CURRENT ETHNOBOTANICAL STATUS OF DISTRICT LODHRAN REGARDING HUMAN BENEFITS FROM APIS DORSATA

### Ahtisham Ahmed<sup>1</sup>, M. Saleem Sabir<sup>1</sup> and Qasim Ali<sup>2</sup>

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Herbal medicines had a distinct position from ancient times till present. Around three-quarter of the world's population depends and lives on old traditional medicinal remedies for their people's basic health treatment. In actual, the shrubs, herbs, or as well plants continuously stay forever human friends. They provide food, shelter and they have helped mankind to heal and cure numerous diseases. Ethnobotany encompasses all kinds of relationships between plants & humans. The concept of ethnobotany may be summed up in four sections, i.e. Humans, shrubs/plants, uses and interactions. Through the previous 100 years ethnobotany studies has progressed and the pattern is changing from documentation approach to a more practical method. Ethnobotany was quite recently been introduced in Pakistan. Regardless of the fact that Pakistan, owing to its diverse climatic and soil conditions, exhibits very rich and unique flora, a few initiatives for documenting plant resources had been launched. The District Lodhran biodiversity was first searched and reported in order to elaborate and organize the accessible dispersed vegetation/plant data and benefits from Apis dorsata. Full ethno-medicinal assessment of 85 species from 43-families identified in Lodhran district was reported and categorized. Research for the survey was carried out between June-2018 and June-2019. The survey research work involved personal analysis, interviews, and plants collection. Specific methods were studied and registered to harvest, dry, storing & process method, and use of medicinal herbs by hakims, local residents and pansaries. The most commonly used therapeutic plants locally against the various illnesses were Coronopus didymus, Withania coagulans, Capparis decidua, Tamarix aphylla, Heliotropium strigosum, Salvadora oleoides and Salsola kali. It was researched that about 74 per cent of the total plant species were therapeutic plants used as ethno-botanical plants within the local surrounding communities.

# EFFICACY OF DIFFERENT FOOD ATTRACTANTS FOR MANAGEMENT OF FRUIT FLIES (DIPTERA: TEPHRITIDAE)

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Fruit Flies (Diptera: Tephritidae) are major pest of different fruits and vegetables in many countries including Pakistan. It is estimated about 55 % losses in fruits and vegetables caused by the attack of fruit flies. Therefore, the

present study was carried out to install the different food substances mixed with Protein hydrolysates for capturing of Fruit flies population in mango orchards. Twelve treatments with three replications were used In T1 (Honey+Bifenthrin), T2 (Sugar+ Bifenthrin), T3 (Banana+ Bifenthrin), T4 (Flour+ Bifenthrin), T5 (Honey+Coragen), T6 (Sugar+ Coragen), T7 (Banana+ Coragen), T8 (Flour+ Coragen) T9 (Honey), T10 (Sugar), T11 (Banana), T12 (Flour). Population data was recorded twice in a week. Our results revealed that T9 (Honey) was most preferred to fruit flies as compared to other Treatments. This approach integrates with other IPM strategies will helpful for management of fruit flies.

# EFFECT OF DIFFERENT NATURAL FOOD ON THE LIFE TABLE PARAMETERS OF COCCINELLA SEPTEMPUNCTATA LINN. (COLEOPTERA: COCCINELLIDAE)

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The present studies were carried out to determine the effect of food on the life table parameters of C. septempunctata L. (Coleoptera: Coccinellidae) in the laboratory, Department of Entomology, Sindh Agriculture University, Tando Jam. The adult beetles were collected around university's field area and reared in the laboratory to maintain the culture for experimentation. The life table parameters of C. septempunctata was observed on various aphid species associated with different host plants. Five aphid species i.e., Lipaphis erysimi on mustard, Macrosiphum avenae on barley, Aphis nerii on Nerium, A. nerii on Akk and Uroleucon compositae on safflower were offered to beetle and their growth recorded. The results indicated that the mean incubation period was 2.41 days at temperature of 22.06 ± 2.15°C when C. septempunctata was reared on different host species with non-significant difference. The longest mean larval period was  $17.07 \pm 0.61$  days recorded on A. nerii (Akk) and the shortest  $9.23 \pm$ 0.60 days on L. ervsimi. The pupal period was 3.07±0.60 days on L. ervsimi. The highest adult male and female longevities were  $36.09 \pm 1.49$  days and  $44.33 \pm 1.69$  days on L. erysimi and the lowest  $16.25 \pm 0.31$  days on A.nerri (Akk). The longest duration of life cycle was recorded 57.28 days on *L. erysimi*; meanwhile, the shortest 36.47 days on A. nerii (Akk), respectively. The highest adult female body weight was recorded 31.75 ± 2.26 mg on L. erysimi and the lowest  $19.36 \pm 0.81$  mg on A. nerri (Akk). The highest fecundity was recorded  $565.55 \pm 7.24$  eggs on L. erysimi and the lowest 332.33±5.61 eggs on A. nerii (Akk). The highest net reproductive rate (Ro) of 429.20 was obtained on L. erysimi and the lowest 94.17 on A. nerii (Akk). The intrinsic rate of increase (rm) and the finite rate of increase (e<sup>rm</sup>) were highest 0.167 and 1.181 on L. erysimi). The results were also correlated which showed the positive  $r^2$ =0.71 (p>0.05) between the body weight of female and fecundity on all diets. Thus, the present findings obtained from this research are helpful in mass rearing of C. septempunctata for timely introduction against different species of aphids.

# STATUS OF PULSE BEETLES, CALLASOBRUCHUS SPP. (COLEOPTERA: BRUCHIDAE) IN SINDH, PAKISTAN

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The survey study was carried out in three ecological zones of the Sindh, Pakistan, i.e., Zone one (Ghotki, Sukkur and Larkana districts), zone two (Hyderabad, Mirpur Khas and Shaheed Benazirabad districts) and zone three (Karachi district) during March – April 2018. Grain samples of major pulses i.e., chickpea (channa), field pea

(matar), cowpea (lobia), green lentil (moong) and daal channa were collected and brought to Stored Grain Research Laboratory, Department of Entomology, Faculty of Crop Protection, Sindh Agriculture University, Tandojam for further examination. Three species of pulse beetle i.e., *C. maculatus*, *C. chinensis* and *C. analis* were identified among the surveyed pulses. Among various pulses, the highest populations of adults, eggs, grubs and pupae was recorded from moong, followed by lobia and gram. No population of pulse beetle was recorded from daal channa and matar. Among the species, relatively higher populations of *C. maculatus* was recorded at all survey districts. The highest population of *Callasobruchus* spp. adults, grubs and pupae was recorded at Mirpur khas district, whereas, the maximum egg population was observed at Sukkur district. Overall among pulses, maximum population of pulse beetles was recorded on mung, whereas, sampling locations of Mirpur Khas district showed the maximum population. Therefore, proper storage and quarantine measures should be taken in the transportation and storing of pulses adequately to restrict the spread and damage of pulse beetles.

#### ROLE OF URBAN PARKS IN CONSERVING BIODIVERSITY OF INSECT POLLINATORS

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Insect pollinators are an important part of the ecosystem as they pollinate maximum food crops consumed by human beings. Without the pollinators, world cannot get enough food. Worldwide, bees have been reported as most efficient pollinators. These little creatures depend upon floral resources to survive. The increasing human activities in the urban areas become a cause of specie decline. Anthropogenic activities such as development of lands for human habitat, electromagnetic pollution and excessive use of pesticides and other chemicals have been reported that harm the bees. Flower visitations rates are affected by the habitat loss due to urbanization. Pollinators especially bees are abundant at those places where there are more flowers. Urban parks not only serve as the recreational spots for human beings but also help in providing more floral resources for insect pollinators. Parks and other green areas help in preserving the insect diversity in urban areas by providing them nesting resources. Positive correlation of bee visitation has been reported before. The parks and green areas which are rich in flowers have more abundance of insect pollinators. Previously, the bees from family Halictidae, Andrenidae, Apidae, Colletidae, Melittidae have been reported in urban parks. Urban parks also support pollination in peri-urban areas and help to sustain their yield.

# IMPACT EVALUATION OF SOME ANTIBIOTICS ON GROWTH, COCOON PRODUCTION AND BACTERIA ASSOCIATED WITH SILKWORM GUT Asma Javaid, Tahir Iqbal and Mubashar Hussain

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Impact of antibiotics on growth, cocoon production and bacteria associated with silkworm gut was assessed in spring 2018. The eggs of Chinese race of silkworm were incubated at standard conditions of temperature (24 - 26 °C), humidity (75  $\pm$  5 % RH) and photoperiod (16 hrs light: 08 hrs darkness). Larval rearing was maintained at recommended conditions of temperature and humidity. Silkworm larvae showing abnormal symptoms were collected and dissected for gut collection. Bacteria were isolated from the gut content by spreading on agar plates and incubated at 37 °C for 48 hrs. Bacterial identification and phylogenetic analysis were carried out by 16S rRNA gene sequencing. The isolated bacteria were subjected to antimicrobial susceptibility test (disc diffusion methods) by using Penicillin (10  $\mu$ g/ml), Tetracycline (30  $\mu$ g/ml), Amoxicillin (25  $\mu$ g/ml), Ampicillin (10  $\mu$ g/ml) and Erythromycin (15  $\mu$ g/ml). Significantly higher larval weight (6.88g), larval growth (5.84cm) and cocoon weight (1.33g) were recorded for larvae fed on leaves treated with penicillin as compared to other antibiotics. All isolated strains showed positive results for the catalase test. We isolated and identified bacterial strains (n = 06) from gut of healthy and diseased

silkworm larvae. Based on 16S rRNA gene sequence, isolated bacteria showed close relation with *Serratia*, *Bacillus* and *Pseudomonas* spp. Notably, 83.3% strains were resistant to Penicillin, Tetracycline, Amoxicillin, Ampicillin and Erythromycin but 16.6% showed antibiotic susceptibility to the above mentioned commonly used antibiotics. Silkworm larvae fed on penicillin treated leaves showed significant improvement in larval weight, larval length and cocoon production. Isolated bacterial strains showed close relation with *Serratia* spp., *Bacillus* spp. and *Pseudomonas* spp.

#### DIVERSITY AND EFFECTIVENESS OF POLLINATORS IN SUNFLOWER CROP

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Sunflower attained 4<sup>th</sup> largest grown crop at worldwide for edible purpose. It contains 35-55% oil material. Pollinators visit flowers to collect pollen and nectars. Pollinators play a critical role in seed production. Cultivated area for pollinators' dependent crops is increasing since last 50 years. Same is true for sunflower the area of which has been increased 358%. Pollinator dependent crops are important part of human diets. Insect pollinators are only responsible for the improvement of cross pollinated crops. In Hymenopterans, honeybees show maximum activity (65.22%) whereas presence of non-Apis bees and scolid wasps has been reported as 20.39% and 1.69%, respectively in sunflower. Wild honeybees show maximum presence for pollination (90.27%) in sunflower. Most of insect pollinators that pollinate sunflower belong to order Hymenoptera, Diptera, Lepidoptera and Coleoptera. Self pollination has also reported in different sunflower varieties. Honeybee activity affects foraging behavior of bumblebees in the same crop. Production of sunflower seed in open flowers is reported to 26% increase as compared to that of bagged ones. However, pollination visitation rate and self fertility changes across different hybrids in sunflower.

### COMPARISON OF THREE LARVAL DIETS FOR REARING ANOPHELES, CULEX AND AEDES SPECIES

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Dengue and Malaria are leading causes of morbidity and mortality in the world including Pakistan. Although significant efforts and resources have been devoted to malaria and dengue control, still there are 3.2 billion people living in the high risk areas all over the world. In Pakistan, malaria is endemic and dengue is spreading to the new areas in all four provinces. Owing to the urging needs for research and development and personnel capacity building for mosquito rearing and training of students and entomologists on vectors control; we at NIFA have developed low cost mixture larval diet for laboratory culture and experiments with Anopheles. The diet has gained worldwide acceptance and need for its refined formulation and rearing efficacy on other mosquito species. The current studies report on the performance of this diet and other related diets on several mosquito species from Culex, Aedes and Anopheles. Accordingly, this diet and its modified formulation was tested against International Atomic Energy Agency (IAEA) recommended diet for mass rearing of mosquitoes. All three diets were tested for growth and development of three type mosquitoes belonging to Anopheles, Culex and Aedes. Each diet was replicated five times and 640 µl of % of each diet was supplied daily to the newly hatched larvae till their emergence to the adult stage. Larval developmental from 1st instar to pupa and to adult and their survival was recorded. Results indicated shortest

larval duration, survival rate from 1st instar to pupa and high emergence rate for NIFA modified diet followed by NIFA diet and finally the IAEA larval diet. Based on its performance, the NIFA modified diet is recommended for Culex, Anopheles and Aedes rearing. Further studies on fecundity and matting competition all three species mosquitoes reared on this diet is warranted.

### BIOLOGY AND FEEDING PREFERENCE OF *TRIBOLIUM CASTANEUM* (COLEOPTERA: TENEBRIONIDAE) IN DIFFERENT WHEAT VARIETIES

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An experimental study was conducted in rearing laboratory, MNS-University of Agriculture, Multan to check the biology and feeding preference of *Tribolium castaneum* on different wheat varieties during 2018. The study revealed that the maximum no. of eggs were recorded in Sehar-06 i.e. 243.54 followed by Shafaq-06, Lasani-08 and Faisalabad-08 i.e. 201, 187.90 and 170, respectively. The maximum development time was observed in Faisalabad-08 (53.01 days), Lasani08 (45.32 days), Shafaq-06 (37.54 days) and Sehar-06 (33.70 days). Biotic potential of *T. castaneum* in different wheat varieties were Sehar-06(0.0712), Shafaq-06(0.0614), Lasani-08 (0.0507) and Faisalabad-08(0.0430). The intrinsic rate of increase of *T. castaneum* in different wheat varieties were Sehar-06 (0.1070), Shafaq-06(0.1032), Lasani-08 (0.08656) and Faisalabad-08(0.07952). was observed.

# DETECTING BIOMAGNIFICATION OF ARSENIC METAL IN FOOD CHAIN OF INSECTS UNDER SEWAGE IRRIGATED ECOSYSTEM IN MULTAN, PAKISTAN

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Vegetables are well thought-out essential for balanced diet since these provide vitamins, minerals, dietary fiber, and photochemicals. Industrialization and urbanization have resulted in accumulation of heavy metals in vegetables grown in peri-urban areas due to accumulation of heavy metals cultivated in the usage of sewage waste water. From Punjab, previous studies have reported accumulation of heavy metals in water, soil and vegetables but none of these studies have evaluated these heavy metals in different insects (i.e. insects pests, predators, pollinators) found in this vegetable agro-ecosystem. Therefore current study is planned to investigate the accumulation of arsenic from vegetables grown in peri-urban areas of Multan. Different vegetables fields were selected that using sewage water. From these fields, samples of insect pests, pollinators and predators were collected along with the samples of water, plant, and soil. These samples were analyzed by using the atomic absorption in order to quantify the arsenic metal from samples by the standards assigned procedure. It was found that arsenic was present in samples of insects pests, predators and pollinators up to varying extent. Future studies should further investigate the impact of heavy metals on beneficial insects i.e. predators and pollinators.

# DESCRIPTION OF THREE NEW SPECIES OF *TETRASPINUS* (ACARI: ERIOPHYIDAE) FROM ZHEJIANG, CHINA

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Eriophyoid (four legged) mites are the most diverse group and many of eriophyoid species are considered economically important pests in agriculture and forestry all over the world. These mites cause not only direct damage to the crops but also transmit viral diseases to the plants that are potentially much more serious losses to the crop than direct damage. These mites are very minute, difficult to see with naked eye but causing heavy losses to cultivated crops. Up to now, the genus Tetraspinus holds 12 species worldwide, of which 8 species were reported from China. 'A survey was carried out to discover the eriophyid mite fauna of Zhejiang Province China. For the collection of eriophyoid mites, Southeast part of China were surveyed during 2015. Different host plants were observed and mites were collected with the help of 30X hand-lens. After collection specimens were preserved in vials having 75% ethanol along with their host plant parts and brought into laboratory for identification. Host plant parts were also kept separately in plant specimen folder for further identification. The details of specimens were labeled on each vial. Keifer's F-medium and modified berlese medium were used for slide mounting. The terminology for the description of body parts of the mites were followed by (Lindquist 1996). Measurements of specimens were followed by (de Lillo et al. 2010) and generic classification was followed by (Amrine et al. 2003). The genus Tetraspinus was recognized by Boczek, 1961 on the bases of type species Tetraspinus lentus (Boczek 1961). Boczek described this genus as; mites belonging to the genus have broad dorsal abdominal furrow and a pair of distinct spines, anteriorly on frontal lobe. The genus Tetraspinus differ from the genus Tetra in having a pair of distinct spines anteriorly on frontal lobe, projecting forward. During present studies, three new leaf vagrant eriophyoid mite species in the genus Tetraspinus were found from Zhejiang Province, China, namely Tetraspinus bungeanus sp. nov. from Euonymus bungeanus (Celastraceae), Tetraspinus Lishuiensis sp. nov. from Bambusa sp. (Gramineae) and Tetraspinus henryiensis sp. nov. from Ormosia henryi Prain (Leguminosae). All described new species in the present studies were found vagrants on lower surface of the leaves, causing no clear damage to their host plants.

# BEHAVIORAL STUDY OF *AEDES AEGYPTI* AND *CULEX QUINQUEFASCIATUS* FOR REDUCTION IN OVIPOSITION ON DIFFERENT WATER SOURCES

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A mosquito belongs to class Insecta and family Culicidae contains 3500 species of mosquitoes, e.g. Aedes aegypti and culexquinquefasciatus. Both Aedes and Culex spp. are potential source of Dengue, Yellow, Fever, Zika infection and Malaria. To control their population by different chemical methods are used but scientists believe to familiarize eco-friendly methods to overcome the serious issues of mosquito due to some biased sorts like species-specific and high environmental hazards. To defeat the issues, a predatory fish tilapia has significance to overcome the population of mosquito. Mosquito Oviposition in the presence of predatory fish may be reduced. In present study, results depict that maximum oviposition was observed in Aedes and Culex larval water source and minimum oviposition was observed in insecticidal water source. The Oviposition significantly reduces accordingly: water source contains Aedes and Culexwater> predatory fish 7-day old Water > predatory fish 10-dayold Water>Aedes and culex water> distilled water> insecticidal water. The result showed that predatory habitat tilapia fish and insecticide application water source have significant impact on the Oviposition of Aedes aegypti and Culexquinquefasciatus.

# CHECKING MOVEMENT OF TICKS (ORDER: ACARI) TOWARDS INFRARED RADIATIONS USED AS A HEAT SOURCE FOR CONTROLLING OF TICK'S POPULATION RATE

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Ticks belonging to the order Acari resembling with mites and considered as major vector for spreading vector borne disease in animals and humans. Alike the insects, ticks having a sensing organ called Haller's organ which help to mediate them towards their host. Present experiment was conducted in the lab of MNS-University of Agriculture Multan to check the movement either ticks use their Haller's organ to sense the infrared lights and show attractive behavior or go away under the lab conditions. Three ranges of IR radiations including 780nm, 880nm and 940nm were selected and used as a heat source during the experiment. Investigations revealed that ticks are phototactic and showed the positively response towards these radiations. Maximally significant response was shown at the infrared range of 880nm. At the IR range of 940nm a large amount of ticks showed an attractive response but comparatively lowest with 880nm. The least significant movement of assessed ticks were shown at the IR range of 780nm. Amongst the three heat ranges the significant range 880nm was observed at which maximum population of assed ticks shown attractive response comparatively other ranges. So, from the above study it is concluded that infrared radiations will be used as heat source for controlling their population instead of chemical usage.

# BIODIVERSITY AND BIOGEOGRAPHY OF SPIDERS IN SUGARCANE FIELDS IN DISTRICT MATIARI & HYDERABAD, SINDH, PAKISTAN

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Biodiversity refers to all the kinds of living things, occupying, living on this globe counting from kingdom Monera to kingdom animalia, undomesticated plants fauna and animals' fauna, micro-organisms, reclaimed or tame animals and plants fauna. The learning of different entities, kinds of insect's and allies' fauna is of immense significance for the reason that supplementary number of the globally reported animals are include spiders. Biogeography is the learning of ways and means by which all the living beings disperse, their process of allocation in. Spider fauna is a carnivorous cluster of terrestrial arthropods and associated with different crop systems including sugarcane crop. Regarding importance of spider's predatory role the two districts were selected and 2345 specimens were collected in 2017 & 2018 during the months May-November. The collected objects were sorted out into nine families and fourteen genera. This loom recommends two guilds of spiders, one on the basis of spiders feeding habits and other on their circadian behaviour. As recent studies have shown assemblages of spiders, impact on pest populations and a better understanding of spider community structure among crops which is crucial in future studies of the arthropod fauna in agro ecosystems.

# SURVEY, SPECIES RICHNESS AND MORPHOMETRIC OF OOTHECAE OF PRAYING MANTIDS IN DISTRICT MIRPURKHAS, SINDH

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Study was undertaken to see the stability and morphometrics of oothecae of the Praying Mantis (Mantodea) which is a fascinating small group of bulky insect and has been superbly engineered for speed and power and has voracious appetite for many type of living things *i.e* arthropods, birds, reptiles, etc. This insect group occurs in warmer parts of the world and reported 436 genera and 2370 species. Reported work from this area is restricted only upon taxonomic along with the biodiversity and biogeography of mantids from Sindh. Due to the little exploration their bio-control agent aspect is the reason to conduct a research in District Mirpurkhas, Sindh (during May to Oct 2018). Total 319 specimens were collected and arranged into 06 genera and 08 species. The morphometrics of identified 49 oothecae were sorted and few healthy were allowed for hatching in fields and at the advanced research laboratory IBGE.

# MOSQUITO FAUNA (DIPTERA: CULICIDAE) OF DIR LOWER, KHYBER PAKHTUNKHWA, PAKISTAN

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Mosquitoes are potential vectors of important human and veterinary diseases. To cope with the risks caused by mosquitoes, evaluation of their diversity, distribution and relative abundance has always been a hotspot of research in diverse habitats around the globe. The present study was aimed to evaluate the mosquito fauna (Diptera: Culicidae) of Dir Lower, Khyber Pakhtunkhwa, Pakistan. Collection of samples was carried out from different outdoor and indoor habitats of Dir Lower during 2018 and 2019. Collection of adult mosquitoes was mostly made through light traps in homes, gardens and animal shelters for nocturnal species. Crepuscular and diurnal species were collected during dawn, dusk and day time from various habitats with the help of insect nets. The specimens were labelled during collection, preserved and mounted with the help of entomological pins in entomological boxes at the Entomology Laboratory, Department of Zoology, University of Malakand. The collected specimens were identified with the help of taxonomic keys of mosquitoes. The collected specimens belonged to three genera namely Culex, Anopheles and Aedes consisting of fourteen species of mosquitoes. Culex quinquefasciatus was the most abundant species with relative abundance of 16.5 % and distribution of 100 % indicating as constant in the study area. An. annularis was having 0.9 % relative abundance (RA) indicating as a satellite species with distribution (C) value of 17 % representing as sporadic species in the study area. The present study encompasses diversity, distribution and relative abundance of mosquito fauna of Dir Lower, KP, Pakistan, indicating the current status of mosquito fauna and associated health risks to veterinary and human population of the study area.

# INSECT FAUNAL DIVERSITY FROM *BRASSICA* CROP ALONG WITH METALS ACCUMULATION (CHROMIUM AND NICKEL) IN THEM IN DISTRICT SIALKOT

### Rafia Ashraf, Sajida Mushtaq, Sadia Maalik, Moazma Batool and Huma Naz

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Sialkot is an industrial city which is well known for sports goods, surgical instruments, leather production and food ceramics throughout the world. Agricultural development, rapid industrialization and urbanization are going towards peak from last ten years. This study was carried out for comparative analysis of diversity of insect fauna

collected from Brassica crop cultivated both in tannery and non-tannery area of Sialkot. Field survey was done, and sampling for data of insect fauna from November 2018 to February 2019. Shannon Diversity index was calculated by comparing the samples of insects from both selected areas. Faunal diversity of insect fauna sampled from non-tannery area was greater as compare to tannery area. Metals accumulation of Chromium and Nickel was recorded in different trophic levels as Brassica crop and insect fauna found on these crops. It well as was determined by Atomic Absorption Spectrometry. Water and soil quality analysis were also done. The results showed that comparatively more metals accumulation was recorded in Brassica crop plant and insect fauna of tannery area as compare to non-tannery area.

# EGG CANNIBALISM AND BEHAVIORAL RESPONSE OF *CHRYSOPERLA CARNEA* (STEPHENS) (NEUROPTERA: CHRYSOPIDAE) IN CAPTIVITY

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Cannibalism is a common occurrence in nature, and many cannibals prey on relatively small and defenseless life stages, such as eggs or young juveniles. Such behavior provides many benefits to the cannibal, but cannibalistic individuals also face risks, including the cost of decreasing their inclusive fitness by eating close relatives such as siblings or offspring. We examined the role of the presence of conspecific larvae of *Chrysoperla carnea* in informing adaptive cannibalism decisions. Larvae of *C. carnea* express low egg cannibalism when alone but become much more cannibalistic in the presence of conspecific larvae and discriminate between their own eggs and those of other females. An experiment also showed that *C. carnea* larvae committed stronger egg cannibalism response on conspecific eggs than did on heterospecific eggs. These results suggest that *C. carnea* larvae become cannibalistic in the presence of conspecifics because they interpret conspecific presence primarily as an indication of decreased likelihood of committing filial cannibalism, and less so as an indication of lower expected survival of eggs or future resource competition. Behavioral locomotion bioassay was video recorded and analyzed using video tracking software (AMY-maze 6.1, Stoelting CO, USA) to examine their preferences during cannibalism. Our study highlights the importance of informational cues, in this case the presence of conspecifics, in modulating the expression of cannibalism.

# ABSTRACTS PRESENTED AT THE FIRST VIRTUAL CONGRESS OF ZOOLOGY

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### **SECTION – III**

### ENTOMOLOGY

# CLADISTIC ANALYSIS OF CHEWING LICE (AMBLYCERA: MENOPONIDAE) INFESTING FOWLS OF DISTRICT HYDERABAD, SINDH, PAKISTAN

### Farheen Shaikh\*, Saima Naz and Nadir Ali Birmani

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The present research work covers the phylogentic relationship of five species which belongs to a family Menoponidae (Amblycera: Phithraptera), recovered from variety of fowls as host from district Hyderabad, Sindh Pakistan. The species are analyzed caldistically by cladogram, using of their apomorphic characters. The Key to the five species of family Menoponidae has also been developed for the three genera. The cladistic relationship of chewing lice species was analyzed to understand the evolutionary aspect of their morphologies as well as their host specificity by using the key characters of lice. This is the first investigation of this kind on various types and breeds of fowls of family Menoponidae from district Hyderabad, Sindh, Pakistan, made a valuable contribution to the chewing lice fauna of Pakistan.

#### ABUNDANCE OF APHIDOPHAGUS HOVER FLIES IN LARKANA, SINDH, PAKISTAN

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Hover flies commonly known as blossom flies fit to large family of small to large flies worldwide about 6000 species have been reported, Aphidophagous hover flies deliver vital ecosystem services as pollinators, biological control agent. Aphidophagous hover flies are one of the important group of flies because their larvae feed on numerous insect pest such as aphids. Jassids, thrips, which feed on various crops like brassica, spinach, rice, wheat etc. present study help in pest controlling of crops in Mohan-jo-Daro Larkana and its peripheral areas. Larkana is the main agricultural area of Sindh where different flowering crops, vegetables and fruits are produced such as Mango, Banana, Guava, Brassica, and Wheat. The present study was conducted from October to, December 2021 from Mohen-jo-Daro Larkana in which different fields were visited during seasonal crops. Total 221 specimens of 2 species (*Episyrphus balteatus*, *Bacha baltetus*) belonging to genus *Episyrphus* and *Bacha* family syrphinae were collected from brassica, spinach and chili paper mostly in aphid colonies.

# STUDY ON THE MORPHOLOGICAL IDENTIFICATION OF FAMILY CHRYSOMELIDAE (COLEOPTERA) FROM DISTRICT BADIN SINDH

#### Sidra Tul Muntha, Naheed Baloch and Riffat Sultana

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The present study was carried out from the district Badin during the year 2021. A total of 432 specimen were collected from the different localities of District Badin and its adjoining areas, and sort out into single family i.e.

Chrysomelidae with only one species i.e *chrysolina gramania*(*Linnaeus*, 1758) it overall percentage was noted 52.08% for Female followed by 47.91% for Male. Collection was made on mint plant belongs to Laminacea family. Present study suggested more in frequent survey in other localities and other host plants.

# A NEW RECORD OF LONG HORNED GRASSHOPPER HEXACENTRUS UNICOLOR SERVILLI 1831 (HEXACENTRINAE: TETTIGONIDAE: ORTHOPTERA) FROM TAULKA DADU

# Asif Nazeer Memon\*, Naheed Baloch, Asif Riffat Sultana, Abdul Sattar, Shamsher Unar, Sidratul Muntaha and Zaryab Gull

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During the preliminary survey of major cropping areas of Dadu a few species of *Hexacentrus unicolor* survilli 1831 were captured which seems a new record for the region. This species is very unique in its morphological appearance and collection of this specimen from Wheat crops is also surprising, this study will conform weather it will proved an major threat the various crops in future or not.

#### TAXONOMICAL SURVEY OF ORB WEB SPIDERS FROM WHEAT CROP, DISTRICT DADU, SINDH

Ali Raza Soomro\*, Tahira Jabeen Ursani, Samina Malik, Jawaid A Khokhar & Asif Raza

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The district gets its name from its headquarters town Dadu. It lies between 24-57and 27-27 north degrees and 67 - 09 to 68 - 25 east longitudes. In Sindh 3.5 million tons wheat is made and Ranked at second number in Pakistan (ESP, 2015-16). Wheat is invaded with number of disturbances, for instance, aphids, caterpillars, crickets, wireworms, leaf bugs, grasshoppers, etc. Farmers generally use pesticides which are harmful for wheat and environment. During the present assessment, wheat field of Dadu was outlined in the time of November, 2019 to April, 2020. 527 models were assembled and managed into two families specifically Araneidae, Tetragnathidae. All families were gathered up to Genera and species level. *A. trifasciata*, *A. Pradhani* of family Araneidae, Tetragnathajavana of family Tetragnathidae are first time recorded from this zone. Unmistakable confirmation of given kinds of different families up to ordinary level similarly as species level with the help of taxonomical keys.

#### FORAGING OF ANTS (FORMICIDAE) INHABITING ON MANGO TREES IN DISTRICT SANGHAR

Asif Raza Soomro, Tahira Jabeen Ursani, Jawaid A Khokhar and Ali Raza Soomro\*

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This research was based on feeding strategies of ants (Formicidae), started from the year April 2019 to December, 2021. Total 7245 specimens were collected and sorted out into fifteen species, and six genera. It was found that ants have different feeding approaches towards the versatile sources of food including insects, small animals, plants, seeds, nectar etc. this co existence have great importance in the Mango fields. Because it was noticed that their role is vital not only for mango fields but for our environment. Hence they are known as sustainers of our environment.

# PREVALENCE OF HEAD LICE INFESTATION AND ITS ASSOCIATED RISK FACTOR IN PRIMARY SCHOOL CHILDREN OF LAHORE, PAKISTAN

### Asma Abdul Latif, Tooba Arooj, Tehreem Asif and Shafaq Fatima

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Head lice infestation is a frequent public health condition that affects people of all ages, but is more prevalent among school-children. The goal of this study was to find out how common head lice infestation in among schoolchildren (age 4-11 years) and their associated risk factors. This cross-sectional study was conducted in different schools of Lahore, Pakistan, from February to May in 2016. A standard questionnaire was design for recording information about risk factors. Visual inspection and combing of hair for 3-4 minutes were made to check the presence of lice, nymphs and eggs. Significant difference for pediculosis and its related risk variables was determined using the  $X^2$  test. There were 141 boys (38.21%) and 228 girls (61.79%) among the 369 students. A total of 243 students tested positive for head lice, resulting in a prevalence of 65.85%. The prevalence for boys were 24.69% and for girls were 75.31%. It was more for girls with significant statistical difference ( $X^2 = 59.095$ , P < 0.001). Prevalence was higher in 6-7 and 8-9 age groups for both sexes. This finding revealed a link between pediculosis and the number of siblings, low-income group, low personal hygiene practices, regularly checked by parents and sharing of accessories. Father's education and mother's occupation showed association with head lice infestation. Pediculosis was found to be inversely related to hair length, dandruff and lubrication of hair. There is a need to raise awareness about the dangers of head lice infestation and to educate teachers and parents on how to avoid pediculosis.

### SOME NEW RECORDS OF EARWIGS (DERMAPTERA) FROM DISTRICT MATIARY SINDH

### Naheed Baloch\*, Haseena Saund, Zainul Abddin, Shamsher Ali and Sidratul Muntaha

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Earwigs belong to order dermaptera are considerable importance. It is important to identify them accurately so that diagnosis of economic problem can be properly made. The Earwigs have been recorded as a pest of Irish sweet potatoes in storage damaging the root of vegetables grown in green houses. As a result of present investigation there are 302 specimens were collected, following species are recorded for the first time from Matiary districts different fields Forcipulla akbari, Forcipula quadrinospinosa, Nala basilis Anisolobis martima, Euborellia anulipus and Anechura fedtschenkoi calciatti.

# EFFECT OF SELENIUM ON POLLINATION EFFICIENCY AND TRIFOLIUM ALEXANDRINUM L. SEED YIELD

### Muhammad Awais Ahmad, Mudssar Ali and Shafqat Saeed

Institute of Plant Protection, MNS University of Agriculture Multan \*Corresponding Author: mudssar.ali@mnsuam.edu.pk

Selenium deficiency may cause mastitis disease in cattle however, by adding selenium in fodder crop it will overcome the disease in livestock. Furthermore, selenium should be added in our staple food as selenium supplement may help human in overcome heart diseases, baldness and sometime cancer. Low dose of Selenium may favor the pollinators, it may extend the life of the bees and also increases the immune and antioxidant activities of the bees. The purpose of our study is to assess effect of selenium on efficacy of different native insect pollinators and ultimate effect of selenium on yield of berseem. There were five treatments of Selenium dose i.e. 00 g/ha (control), 05 g/ha, 10 g/ha, 15 g/ha and 20 g/ha and two sprays of Selenium were applied at one week interval. In our experiment there were three dominant groups of pollinators solitary bees (*P. oxybeloides, Xylocopa sp.*), Honeybees (*A. dorsata, A. mellifera, A. florea*) and syrphid flies (*E. aeneus, E. balteatus, I. scutellaris*) were found visiting berseem crop.

Moreover, our study suggest that high dose of selenium in plant cause decrease in abundance, visitation rate and visit duration of pollinators however, low dose of selenium (5g ha<sup>-1</sup>) assist pollination. In our study, maximum seed yield (number of seed per head, seed weight per head, 1000 seed weight) of berseem was obtained at low dose of selenium (5g ha<sup>-1</sup>) On the other hand plant height and head weight were not affected by high dose of selenium, however, reduction in biomass of plant was observed at high dose treatments (15g ha<sup>-1</sup> and 20g ha<sup>-1</sup>). By applying suitable dose of selenium to the plants, may increase the seed yield as well as it will favor the pollinators.

#### THE OCCURRENCE OF GYNANDROMORPHISM IN BEES (HYMENOPTERA, APIDAE); A REVIEW

### Danyal Haider Khan, Mudssar Ali, Muhammad Awais Ahmad, Shafqat Saeed

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Gynandromorphism is an abnormal phenomenon by which both male and female phenotypical characters appear in an organism. Genetic mutations in the sex determining genes were thought to be the cause of this abnormality but latter it was reported that rather of being induced by genetic mutations, some anomalies in the preservation of regulatory signals that govern sex differentiation at single cell lineages cause it. There are basically three forms of gynandromorphism, bilaterally asymmetrical, mosaic and transverse type. The first one happens at early stages of development, when an organism's body has one side that is male and the other side that is female, the second one occurs latter in development, when the two sex characters are not clearly defined, whereas last one occurs when two asymmetrical portions of the sex characters are distributed. Gynandromorphism is reported in 13 orders and 69 families of insects till now. In bees it is documented in 140 species from all main biogeographic areas of the world, representing 35 genera from all families of Apoidea. The bees in the genera Megachile and Xylocopa have the most gynandromorphs documented. Females are more likely than males to experience cross-sex manifestation of character states. The purpose of studying gynandromorphism is to observe the role of sex linkages in evolution of a species.

#### **Diversity of Pollinators in Oil Seeds Crops**

### Shahid Iqbal\*, Mudssar Ali, Danyal Haider Khan and Muhammad Zubair

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Pollination is a very essential service for the ecosystem in terms of maintaining the biodiversity of the important plants on earth. It contributes toward 35% of crop production all over the globe. Insect pollinators especially Honey bees enhance the seed weight, filled seed per head and seed yield per head in oilseeds crops and increase 30% of seed production. Pakistan imports 1.98 MT edible crude oil by spending about 45 billion rupees. The relative abundance of insect pollinators has a favorable effect on yield and yield components. About 12 insect visitor's affiliated to different orders Coleoptera, Lepidoptera, Diptera, and Hymenoptera were founded visiting the flower of sunflower head. The abundance of Hymenopterans were greatest then Dipterans and then others insects were founded. In Hymenopterans, honeybees show highest presence (65.22%) although Non-Apis bees and Scolid wasps has been recorded 20.39% and 1.69% respectively. The Hymenoptera order has the highest abundance rate of insect pollinators (89.8%), followed by Diptera (6.7%) and Lepidoptera (6.7%). The oil content and seed yield is increased by 6% and 30% respectively in hybrid varieties of sunflower by pollination through honey bees. Improving the populations of the pollinators improve the yield of oilseeds and other crops. It provide great nesting sites to the bees which help in securing the habitat for them.

#### ROLE OF MANAGED HONEYBEES IN ALFALFA SEED PRODUCTION

#### Kamran Ejaz, Mudssar Ali and Shafqat Saeed

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Alfalfa (Medicago sativa L.) is one of the most important fodder crops having 20-24 % protein content more than other widely used fodder crops and is well adopted to wide range of climates and soils. Due to high yield and nutritional value alfalfa called as queen of fodders. It requires bee pollination for seed production. Alfalfa is a perennial, cross-pollinated crop that requires bees to "trip" flowers in order to release pollen for seed development. Tripping is normally done by bees; Wild bees are effective alfalfa trippers and hence good cross-pollinators. The honey bee worked for as long as 11 hours a day, and pollen-collecting individuals opened 7 flowers in a minute which makes them also important pollinator alfalfa crop. To increase their yield alfalfa, seed farmers rent bees from Beekeepers, who strategically place the hives near to their blooming alfalfa fields to enhance pollination and seed output. Honey bees (Apis mellifera L.) are not enough for alfalfa trippers for efficient seed production in compared to solitary bees. Furthermore, Honeybees are greater in number and easily managed while solitary bees are efficient but hard to conserve. Yields per acre may decline by more than half if honeybees were only used for seed production. The decline may be greater than 90% if honeybees were completely missing. Bee pollinators are responsible for 80% of the world's plants pollination including 90% of different food crops. Solitary bees and honey bees both play an important role in pollination of alfalfa crop and its seed production. By placing honey bee hives in alfalfa field and by conserving solitary bees we can get maximum seed yield of alfalfa.

### EFFECT OF SELENIUM ON POLLINATION EFFICIENCY AND TRIFOLIUM ALEXANDRINUM L. SEED YIELD

#### Muhammad Awais Ahmad, Mudssar Ali and Shafqat Saeed

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Selenium deficiency may cause mastitis disease in cattle however, by adding selenium in fodder crop it will overcome the disease in livestock. Furthermore, selenium should be added in our staple food as selenium supplement may help human in overcome heart diseases, baldness and sometime cancer. Low dose of Selenium may favor the pollinators, it may extend the life of the bees and also increases the immune and antioxidant activities of the bees. The purpose of our study is to assess effect of selenium on efficacy of different native insect pollinators and ultimate effect of selenium on yield of berseem. There were five treatments of Selenium dose i.e. 00 g/ha (control), 05 g/ha, 10 g/ha, 15 g/ha and 20 g/ha and two sprays of Selenium were applied at one week interval. In our experiment there were three dominant groups of pollinators solitary bees (*P. oxybeloides, Xylocopa sp.*), honeybees (*A. dorsata, A. mellifera, A. florea*) and syrphid flies (*E. aeneus, E. balteatus, I. scutellaris*) were found visiting berseem crop. Moreover, our study suggest that high dose of selenium in plant cause decrease in abundance, visitation rate and visit duration of pollinators however, low dose of selenium (5g ha<sup>-1</sup>) assist pollination. In our study, maximum seed yield (number of seed per head, seed weight per head, 1000 seed weight) of berseem was obtained at low dose of selenium (5g ha<sup>-1</sup>) On the other hand plant height and head weight were not affected by high dose of selenium, however, reduction in biomass of plant was observed at high dose treatments (15g ha<sup>-1</sup> and 20g ha<sup>-1</sup>). By applying suitable dose of selenium to the plants, may increase the seed yield as well as it will favor the pollinators.

#### SURVEY OF GENUS GRYLLUS (GRYLLIDAE: ORTHOPTERA) IN SINDH

#### Naila Bhanger, Riffat Sultana, Naheed Baloch and Santosh Kumar\*

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Several species of family Gryllidae occur in different habitats i.e trees, shrubs, herbs and grasses. Its population was high during April, May and June. Rainfall varies from year to year. During present study we have collected about 85 specimens of Genus *Gryllus* and sorted them out into two species *Gryllus* (*Gryllus*) *bimaculatus* De-Geer,1773 and *Gryllus* (*Gryllus*) *campestris* Linnaeus, 1758. Sex was also identified on bases of wings, male have shorter, sturdier wings with rough underside surfaces known as file. These insects are omnivores, scavengers and herbivores. During field surveys male crickets were found dominant and show aggressive behavior towards females and to protect their territory. Several morphological variations were found; present study suggests that it might be due to geographical variation further, detailed surveys are needed.

### OBSERVATION ON OVIPOSITION BEHAVIOR OF *POEKILOCERUS PICTUS*UNDER LABORATORY CONDITIONS

#### Kavita Bai and Riffat Sultana

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Poekilocerus pictus (Fabricius, 1775) is also known as Akk grasshopper or locally in a few tribal areas called Titighodo. It is a principal pest of *Calotropis procera* (Akk), but it also damages wheat, alfalfa, papaya, citrus, castor, brinjal and cow pea on large scale in India as well as in Pakistan. Some observations were made on the reproductive activities of the *P. pictus* under laboratory conditions. On attaining sexual maturity, the males took 3 to 4 days for copulating once final shedding, whereas the females took one to a pair of days or in rare cases sexual practice started instantly once final molt in each sex. After the 2-3 days of copulation females start egg-laying and deposit about 60-80 eggs and 2-3 egg pods during their entire life. It was also pointed out that oviposition normally occurs during the daytime mostly in the morning. Number of eggs reduces with increase in the frequencies of oviposition.

#### ENTOMOPHAGY: DIVERSITY OF INSECTS USED AS A FOOD SOURCE FOR HUMAN BEINGS

#### Muhammad Shafiq\*, Wajid Abbas, Saifullah Shafiq and Umair Siyal

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In 2050 world population is expected to be over 9 billion, to meet people's feed is a major discussion hotspot. The practice of eating edible insects has been recognized for a long time. Finding other sources like insects that have equal nutrients value to fish and meat are highly needed. Over 2300 species of 18 orders are reported as edible insects. Larvae of *Callipogon barbatus*, *Rhynchophorus phoenicis*, *Hermetia illucens*, and pupae of *Musca domestica* are the most consumable edible insects due to their massive protein contents. Insects are rich in crude protein, vitamins, minerals and less in fat and cholesterol. Imago is also preferred like *Poekilocerus pictus*, *Ballatodia*, and *Apis mellifera*. Some insects are well accepted due to their flavor like aquatic insects have fish taste, the termite has a nutty flavor, ants are sweet, and stink bugs have apple flavor. People are using insects as they are inexpensive and even free to consume. For imminent food insecurity, feasible methods and strategies should be implemented to promote the consumption of edible insects.

#### IMPORTANCE OF BLOW FLIES WITH RESPECT TO FORENSIC ENTOMOLOGY; A REVIEW

#### Alina Sabir, Mudassir Ali and Shafqat Saeed

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The Calliphoridae (commonly known as blowflies, flashflies, carrionflies, blue bottles, green bottles, or clusterflies) is a family of insects in the order Diptera, with 1,200 known species. Blowflies are important in forensic entomology, where the maximum and minimum postmortem intervals (PMI) are estimate on the basis of developmental stages of Dipteran larvae and pupae that consume dead tissue. Blowfly is usually the first insect that comes in contact with carrion, because they have the ability to smell dead animal matter from up to 2 km (1 mi) away. Females deposit eggs on carrion. Traditional estimations of time since death are generally recorded, the minimum period of insect (PIAmin: 72 hr) and (PMI:<48hr), assuming colonization occurred after death, blow fly specimens found infesting a corpse are used to determine if the corpse was relocated or if the individual ingested narcotics prior to death. The entomologist must know how the blowfly behaves specifically in the area where the body was discovered. This involves recording environment temperatures, time of day, condition of the body at the crime scene as well as retrieving a history of the climate in the region. *Calliphora livida, Calliphora vicina* and *Cynomya mortuorum* are important flies of forensic entomology. In this study, we determine morphology of all stages, their development and growth and the comparison of life cycle of different *Calliphora livida, Calliphora vicina* and *Cynomya mortuorum*. The results of this study may be helpful in carrion dead time investigation and also the effect of temperature variation on biology of these species.

#### TAXONOMIC STUDIES OF THRIPS TABACI ON ALLIUM CEPA IN PAKISTAN

#### Sadique Ibrahim, Mirza Abdul Qayyum, Saifullah and Unsar Naeem-Ullah

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Onion, Allium cepa is one of major dietary crop in Pakistan with high level of nutrients like Phosphorus, Calcium, and Carbohydrates. Pakistan is 6th largest onion producing country in the world with a share of 3% at international level. Among all vegetables produced in Pakistan, onion ccontribute 40%. Annually, Pakistan exports around \$ 125 million vegetables of which onion, garlic, and leeks has up to \$ 57 million in 2020. There are many reasons of lower production of this crop. Thrips tabaci has become a regular pest all over Pakistan. It causes both direct and indirect damage by feeding and ovipositing on leaves also transmit pathogens that reduced bulb size and quality. Due to it's smaller size cryptic habits, there are several problems in managing it. Th present study is being devised to observe biodiversity of this pest in vegetables. This would help to understand and foresee the pest management of Thrips tabaci for future needs.

## INTERACTIVE EFFECT OF NUTRITION AND POLLINATION ON CROP PRODUCTION- A REVIEW

#### Muhammad Zubair\*, Mudssar Ali, Shafqat Saeed, Shahid Iqbal, and Muhammad Awais Ahmad

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Pollination is necessary for enhancing the crop production because 70% of the crops depends upon insect pollinators. The available nutrients and insect pollination both are necessary for increasing the yield of flowering crops. The relationship between the pollinators and flowering plants is very important for the maintenance of ecosystem services. Flowering plants are main source of insect pollinators because they provide nutrition to the insect pollinators. Most of the studies have been reported variable results for different levels of fertilizers on insect pollination and crop production on different crops i.e. it was found that common bean production was increases due

to insect pollination at low nitrogenous fertilizer level. In Sunflower the maximum yield was reported at intermediate Nitrogen fertilizer level due to enhanced insect pollination. Moreover, in rapeseed the insect pollinator's visitation and crop yield was maximum at high dose of fertilizer (NPK). In alfalfa crop the yield and yield parameters were recorded maximum at the intermediate level of Phosphorus while at low level of phosphorus the crop production was decreased and ultimately pollination decreases. In Sunflower crop the effect of Potassium fertilizer was also reported. The crop production in terms of yield parameters such as head weight, head diameter, seed weight and number of seeds was maximum at the intermediate level of K. The maximum abundance of insect pollinators was also recorded at medium level of Potassium. The plant nutrition and insect pollination was necessary to increase the crop production and these studies would help us to determine best level of fertilizer at which the maximum benefits of insect pollination and seed production can be achieved.

# DOES THE DURATION OF THE PRE OVIPOSITION PERIOD OF HONEYBEE QUEENS AFFECT THE HONEY PRODUCTION OF COLONIES?

#### Muhammad Khalid Rafique\*, and Rashid Mahmood

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Data of the pre oviposition period (POP) of honey bee queens were collected from 25 colonies at mating station at HBRI NARC. Generally, the pre oviposition period of the tested queens lasted from 6 to 34 days with an average of 15.8 days. About 80.1% started egg laying 8 to 18 days after emergence. The length of POP varied significantly among years and months and among different queen bee breeders. Both, the type of mating and the mating location significantly affected POP. Artificially inseminated queens had the significant highest value (17.6 days) of POP in comparison with naturally mated queens at Islamabad (15.4 days) and margallah hills -mating (14.9 days) stations. The relationship between POP and honey production of bee colonies was not found to be significant. The results of this study have demonstrated that honey production, which is assumed to characterize queen vitality, is not affected by POP but that POP is highly affected by several environmental factors during the period before the start of egg laying. Consequently, the pre oviposition period of honeybee queens should not be used as a tool to pre-evaluate honeybee queens.

## COMPARISON BETWEEN THE ATTRACTION OF NATIVE AND NON-NATIVE FLOWERING PLANTS TOWARDS INSECT POLLINATORS

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Biodiversity of the pollinators is decreasing globally due to various human activities i.e. agricultural infestation, habitat loss, pesticide use and low availability of flora. Habitat loss is one of the most critical factor in pollinators declining. Shortage of floral supplies and nesting habitat are among the natural and human-caused stresses that also have contributed to pollinators decline Moreover, managed bees (*Apis mellifera*) are also facing colony collapse disorders worldwide. There are native and non-native flowers were grown to attract the diversity of pollinators. About 90% of the flowering plants provide foraging resources for diversity of insect pollinators. Bees attracted towards the native plant but the solitary bees were attracted towards the non-native plants. Abundance of pollinators was directly proportional to the numbers of blooming flowers. Exotic and near native plants have an important role in enhancing the habitat for pollinators along with the native flowering plants. Temporal stability of floral resources is important for the reproduction of the pollinators but there were not work is done on conservation of insect pollinators by using these flowering plants in Pakistan due to this different bee species were unidentified in Pakistan till now. Hence by providing these floral resources we can conserve the pollinators. We suggest that these flowers should be tested and choose the highly attractive flowering plant of that area because native plants are cheap and easily available.

### MASS REARING OF ACRIDA EXALTATA (ORTHOPRTRA: ACRIDIDAE) UNDER LABORATORY CONDITION

#### Muhammad Younus, Santosh Kumar, Riffat Sultana\*, Muhammad Shahzad and Shamsa Kanwal

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Preliminary investigation was carried out in order to reared *Acrida exaltata* (Walker) under laboratory conditions. Target insects were reared on different diet and food consumption and utilization, growth (growth rate, average daily growth, specific growth rate and wet weight gain), survival and life span were noted. It was found significantly different in various diets. A detailed study is under progress.

## GRASSHOPPERS (ACRIDIDAE: ORTHOPTERA) OF TALUKA KANDIYARO, DISTRICT NAUSHAHERO FEROZE

#### Bahadur Ali Gadani, Waheed Ali Panhwar, Muhammad Furqan, Mohammad Zubair and Abdul Qayoom

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Grasshoppers are the most numerous and diverse insect species. Grasshoppers have many advantages for such experiments, including their large body size, quick capture capacity, and high dominance, to the point that they have become a primary invertebrate category for biological indication in a broader context. They are found in all ecological systems and have a major economic impact due to their ability to destroy nearly all types of green vegetation. In grasslands, they are often the main invertebrate eaters and provide a valuable food source for a number of predators, including birds, lizards, and other mammals. The grasshoppers are classified in two categories: Caelifera which are short horn grasshopper and other are Ensifera which are large horned grasshopper. At present extensive surveys were conducted from taluka Kandiyaro district Naushahero Feroze to collect the grasshoppers. About 170 samples were captured and sorted out into 05 sub- families i-e: Acridinae, Cyrtacanthacridinae, Eyprepocnemidinae, Oxyinae, Spathosterninae and 10 species i-e: Acrida exaltata, Truxalis eximia eximia, Phlaeoba tenebrosa, Anacridium aegyptium, Anacridium rubrispinum, Cyrtacanthacris tatarica, Eyprepocnemis alacris alacris, Oxya velox, Oxya bidentata and Spathosternum prasiniferum. Besides, detail genitalia study is provided first time from the studied region. Definitely, this study will be helpful for the future scholars.

#### STUDY ON DRAGONFLIES OF DISTRICT KHAIRPUR MIRS SINDH PAKISTAN

#### Summaya Naimat Shaikh, Abdul Manan Shaikh and Waheed Ali Panhwar

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Dragonflies and damselflies are part of the order Odonata, which is primarily found in tropical and subtropical climates. Except for the Antarctic region, reports of dragonflies come from everywhere across the planet. A majority of them live in countries with warmer climates or in tropical regions .From March 2021 to August 2021, research was conducted to gather data on the Khairpur district's dragonfly fauna. Two families were represented by a total of 219 specimens, of which nine species were found in five genera. Eight species of Libellulidae are found in four genera, while just one species is found in the Aeshnidae family. Compared to the Aeshnidae, the Libellulidae has the most species. Orthetrum Chrysis Orthetrum sabina Orthetrum pruinosum neglectum Orthetrum cancellatum Acisoma panorpoides Pantla flevescens Crocothemis nigriforns Crocothertumis servilia and Anax imperator were among the species found. the maximum proportion of Libellulidae was found to be 85.0 %, while the lowest percentage of Aeshnidae was found to be in the family Libellulidae 14.65 %.

### SURVEY OF FAMILY GRYLLIDAE FROM VICINITY OF SHAH ABDUL LATIF UNIVERSITY KHAIRPUR

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Gryllidae is the family of order Orthoptera that include insects called Crickets. Fossil evidence show that it belongs to the Jurassic period. In many parts of geographical distribution, populations of crickets have been decreased few decades ago. The crickets are endangered in central and northern Europe. The Gryllidae commonly use their mouthparts to dig out the soil and make a burrow, sometimes, they use their hindlegs for this purpose. Crickets call their partners for matting purpose by rubbing their wings having specialized location in their forewings. During the current study, survey of Gryllidae fauna was carried out from vicinity of Shah Abdul Latif University Khairpur. The collected samples were brought into entomological laboratory Department of Zoology, Shah Abdul Latif University Khairpur. The samples were killed by standardized entomological method and then were stretched on the stretching board. The specimens resulted in finding of 06 species of Gryllidae i.e: *Acheta domesticus*, Linnaeus, 1758, *Acheta meridionalis*, Uvarov, 1921, *Teleogryllus (Macroteleogryllus) mitratus* Burmeister, 1838, *Gryllus bimaculatus* De Geer, 1773, *Gryllodes sigillatus* Walker, 1869 and *Gryllodes supplicans* Walker, 1859 respectively. Additionally, the description of species along with digital images are also provided. Hopefully, this study will prove to be a base line for future researchers dealing with Gryllidae fauna.

#### DOCUMENTATION OF THE ORTHOPTERA FAUNA OF KHYBER PAKHTUNKHWA, PAKISTAN

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The Orthoptera fauna of Pakistan is one of the least explored in Pakistan particularly KhyberPakhtunkhwa province. The geographical conditions of Pakistan are of great importance with its intermingling of oriental, Palaearctic and Afro-tropical elements and prevent an ideal situation for the study of Orthoptera fauna. Unfortunately, information available about the species of Orthoptera and their distribution are quite inadequate. Therefore, present study was designed for documentation of Orthopteran fauna of this region. Present study was conducted from KhyberPakhtunkhwa, Pakistan. As a result of this study, Diabolocatantops innotabilis, Diabolocatantops sp, Anacridium aegyptium, Anacridium rubrispinium, Cyrtacanthacris tatarica, Acrotylus humbertians, Acrotylus longipes longipes, Aiolopus thalassinus tamulus, Truxalis eximia eximia. Oxya velox, Oxya hyla hyla, Oxya bidentata, Acrida exaltata, Phlaeoba tenebrosa, Eyprepocnemis alacris alacris, Spathosternum prasiniferum, Phaneroptera spinosa, Phaneroptera roseata, Trigonocorypha unicolor, Hexacentrus unicolor, Euconocephalus incertus, Euconocephalus pallidus, Euconocephalus nasutus, Euconocephalus mucro, Acheta domesticus, Gryllus bimaculatus were recorded.

### STUDY ON PYRGOMORPHIDAE (ORTHOPTERA) OF TALUKA UBARO DISTRICT GHOTKI SINDH PAKISTAN

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Pyrgomorphidae is a family of the order Orthoptera under the suborder Caelifera. The members of this family are generally known as gaudy grasshoppers. At present surveys were carried to collect the Pyrgomorphidae fauna of taluka Ubaro. Nearly, 240 specimens were collected and sorted into 05 genera viz: *Chrotogonus* Serville, 1838, *Tenuitarsus* Bolívar, 1904, *Pyrgomorpha* Serville, 1838, *Atractomorpha* Saussure, 1862, *Poekilocerus* Serville, 1831

and 11 species i-e: Poekilocerus pictus, Chrotogonus brachypterus, Chrotogonus homalodemus, Chrotogonus oxypterus, Chrotogonus trachypterus, Tenuitarsus orientalis, Pyrgomorpha bispinosa, Pyrgomorpha conica, Pyrgomorpha inaequalipennis, Atractomorpha acutipennis, and Atractomorpha crenulata. The current study focused on conventional morphological features together with a thorough study of genital structures of Pyrgomorphidae for a clear understanding of the purpose of morphological characters. In addition to this, taxonomic keys are also prepared for easily identification the Pyrgomorphidae species.

#### UNDERSTANDING THE ROLE OF ENVIRONMENT IN SANDFLY POPULATION FLUCTUATIONS

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Sandfly belong to the order Diptera and it's becoming a threat for the mankind. More than 1000 species of sandflies has been documented till so far. Among them only, *Phlebotomus* and *Sergentomyia* genera has been recorded to be problematic in terms of disease transmission which is Leishmaniasis. However, disease spread is mostly dependent on the environmental changes. Among the factors, biotic and abiotic factors play an important role in favouring the sandfly population. Thus, to access it, study was designed. Fixed and non-fixed sites were selected to maximise the collection and to collect the samples A4-sized sticky traps were used. Results depicted that, *Phlebotomus alexandr* showed positive correlation with the temperature while the *Phlebotomus major* showed negative correlation with the temperature. This study will help the concerned departments in the management of sandflies.

# DESCRIPTION OF NEW RECORDS ON *BLAUTA CRIBRARIA* (GERMAR, 1844) (COLEOPTERA: ELATERIDAE: CARDIOPHORINAE) FROM SINDH PAKISTAN

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The research was carried out in (2018-2020) from the Ghotki district of Sindh Pakistan. In addition, this studies indicated that the population of new recorded species were considerably different on its geographical distribution of click bettles (Elateridae) fauna of different districts. Identification of new recorded species has been done based on the taxonomical characteristics such as external morphology and male genitalial structures. *Blauta cribraria* near to *Blauta falli*, resembled to in the redish brownish body coloration black in *Blauta cribraria* but in *Blauta falli* dense punctuation on pronotum, but slightly differences, pubescence smaller mandibles. Ventral carina with prosternal spine, scutellum and metacoxal plate with a tooth, rostrum circular and genitalial bases have a small variations in space of aedeagus median lobe.

#### SECTION – I V

#### PARASITOLOGY

### EPIDEMIOLOGY AND RISK FACTOR ANALYSIS OF FASCIOLOSIS IN BUFFALOES IN DISTRICT BAGH

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This research work was carried out in district Bagh, Azad Jammu, and Kashmir, Pakistan, extending from December 2016 to May 2017. This is the first-ever attempt to find out the prevalence of fasciolosis in buffaloes (*Bablus bubalis*) of district Bagh. During this study 200 fecal sample was collected, it was found that 40 percent fecal samples contain eggs of *Fasciola hepatica* and 60 percent fecal samples were negative. Liver samples of slaughtered buffaloes were also examined during the study period and it was found that 74 samples have faciola, out of 200 samples, thus 37 percent of samples contain *F. hepatica* and 63 percent buffaloes liver samples were negative. It was observed that 19.25% of animals of the area have parasites in their bodies, which are causing economic losses to the owners, in the form of less production of milk and meat and more mortality among the livestock and damages to farmers.

# SUITABILITY OF PARASITOID WASP, *BRACON HEBETOR* S. (HYMENOPTERA: BRACONIDAE) AS ECTOPARASITOID OF *SCIRPOPHAGA NIVELLA* F. (LEPEDOPTERA: CRAMBIDAE).

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The Braconid wasp *Bracon habetor* S. (Hymenoptera: Braconidae) is consider as voracious ectoparasitoid of a wide range of Lepidopteran pests including sugarcane top borer (*Scirpophaga nivella*). The suitability of *B. hebetor* was evaluated on *S. nivella* at IPM Lab, College of Agriculture, BZU Bahadur Campus Layyah, Pakistan during 2017-18. *Bracon hebetor* was cultured on the wax moth (Lepedoptera: Galleriinae) larvae which were already reared on honeybee wax in lab conditions. The 3<sup>rd</sup> and 4<sup>th</sup> larval instars of *S. nivella* were collected from insecticidal free experimental sugarcane field plots. Significantly higher number of eggs was found on 4<sup>th</sup> instar (5.25±0.47) on host pest than on 3<sup>rd</sup> instar (3.25±0.47). Similarly, elevated trend of adult emergence (male/female) was also recorded from 4<sup>th</sup> instar of host body. The adult longevity of *B. hebetor* was recorded more on 4<sup>th</sup> instar of *S. nivella* (9.75±0.85) days followed by 5.5±0.64 days on the 3<sup>rd</sup> instar of host insect. The preliminary results revealed that *B. hebetor* is a potential parasitoid and can be included in IPM module for *S. nivella* and for other lepedopterans pests of sugarcane also. However, studies on field augmentation and conservation of this biocontrol agent are also warranted to evaluate its success in agro ecosystem.

### PREVALENCE OF FASCIOLIASIS IN LIVETOCK OF DISTRICT LAKKI MARWAT KHYBER PAKHTUNKHWA

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Fascioliasis is a disease commonly found among cattle, buffalos, sheep, goats, horses, donkeys, rabbits, wild ruminants and humans. The disease is caused mainly by two species of parasitic Trematodes that affect the liver and other associated organs. Liver flukes belong to the group of food borne Trematodes infection and are zoonotic. The main objectives of this study were to determine the prevalence of fascioliasis infections in cattle and buffaloes, slaughtered in Lakki Marwat district slaughterhouses. The slaughtered animals were weakly inspected for liver fascioliasis from November 2018 to April 2019. Fascioliasis was detected from a total of 210 basing on animals specie, sex, season, and locality. The total prevalence rate of Fasciola sp. infection occurs in the study area were about 52/210 (24%) from the total Cattles slaughtered. The fascioliasis level was 35/115 (30%) in cows. In goats 5/45 (11%) were infected with fascioliasis. Sheeps infected were 8/30 (27%) and in buffalos 4/20 (20%) were infected cases of fascioliasis. Moderate level of fascioliasis was observed in Cattles slaughtered in the abattoirs of district Lakki Marwat. The highest level of fascioliasis was observed during the winter. It constitutes a major cause of economic losses at district Lakki Marwat.

# THE HELMINTH PARASITES OF FRESHWATER FISHES IN FERGANA REGION, UZBEKISTAN

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The Fergana Valley of Uzbekistan is rich in water resources - there are a number of piedmont reservoirs (Asaka, Kurgantepa, Karkidon, Rezaksay, Chartak, Eskiyar, etc.) and floodplain lakes (Sarykamysh, Mingbulak, Tudakul, etc.). The valley is indented with say, rivers (Naryn, Karadarya, Syr Darya, etc.), and irrigation canals. In the Fergana Valley there are numerous, of various sizes, isolated bodies of water - ponds, formed naturally or artificially created by people. The successful introduction of fisheries in reservoirs is often limited by parasitological factors. Many parasites cause diseases, and in some cases, the mass death of fish. Accurate knowledge of the parasitological situation in water bodies contributes to the rational introduction of commercial fisheries and increases their productivity. In 2018-2019 years helminthological material was collected of fish on the upper parts of the Sir Darya river, fisheries farms of the Dangara and Besharik districts of Fergana region, as well as the Isfaramsay, Sukhsay, Shohimardonsay, Naimansay rivers, canals of the Big Fergana, Souths, Andijan and the Rishtanabad collector in the territory of Fergana region. By the method of complete and incomplete helminthological autopsy, 347 specimens fish were investigated in all the named water bodies of the Fergana, fishes belonging to 5 species, including, Schizothorax intermedius - 56 pcs., Carassius auratus gibelio - 47, Cyprinus carpio - 68, Pelecus cultratus - 84, Ctenopharyngodon idella - 39, Silurus glanis - 34 and Channa argus warpachowskii - 19. In fish of the studied reservoirs, we found 19 species of parasites belonging to different systematic groups; cestodes - 7 (36.8), trematodes - 5 (26.3%), nematodes - 5 (26.3%) and acanthocephalans - 2 (10.5%). Nine, out of 19 species, are noted in larval form and ten in sexually mature form. The dominant species were the genera of cestodes - Ligula and Digramma, and nematodes - Raphidascaris. The development cycle of these helminthes proceeds with the participation of the first (oligochaetes, copepods, and many other groups of invertebrates), the second (dragonfly larvae, caddis flies, beetles, benthos-eating carp fish) and definitive hosts - predatory fish. The taxonomic diversity of cyprinid fish parasites in the region under study is represented by rich fauna of cestodes, trematodes, nematodes, and slightly acantocephals.

## DISTRIBUTION AND POPULATION DENSITY OF TERRESTRIAL MOLLUSKS INTERMEDIATE HOSTS HELMINTS IN THE FERGHANA VALLEY

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These mollusks are considered the intermediate host of many helminths, and in their reproduction plants play an important role, first of all, they are used as nutrients from plants, and most of them feed on the green part of the plant, a certain part and the humus cover of the plant. Secondly, the vegetation cover is their habitat, as many mollusks use them as a shelter in adverse weather conditions, and for a certain species it is considered a breeding place. Therefore, we obtained data on the prevalence of mollusks and their population density depending on their prevalence in high-altitude areas of plants. According to the results of the study, 9 species of terrestrial mollusks are distributed in the steppe region of the Ferghana valley, which are involved in the spread of helminth diseases in pastures, the degree of density of which in their populations is different. Entering a type with a high population density, Xeropicta candacharica can meet it on an area of 1 m.sq. with an area of more than 100 pieces. The type where the degree of density is average is Pupilla muscorum, it is from 50 to 100 pieces in 1 m.sq. The density level is from medium to low species, the density of their population, included in the species Cochlicopa nitens, C.lubrica, Vallonia costata, Bradybaena dichrozona, Leucozonella mesoleuca, Angiomphalia regeliana, Bradybaena lantzi, Deroceras laeve, is from 1 m.sq. to 5 soles, their density is from 5 to 50 soles, from 1 m.sq to 5 soles, in low species Zonitoides nitidus is from 1 m.sq. to 5 soles. In the area of Adyr, it was found that the intermediate host of helminths is the distribution of terrestrial mollusks and the density in the population (per 1 m.sq.) is as follows: Cochlicopa nitens (15-17 of piece), C. lubrica (4-6 of piece), Vallonia costata (14-15 of piece), Pupilla muscorum (15-18 of piece), Pseudonapaeus albiplicataus (9-10 of piece), Ps. sogdianus (11-12 of piece), Gibbulinopsis signata (8-12 of piece), Turanena tenuisira (7-9 of piece), Br. phaeozona (3-4 of piece), Leucozonella mesoleuca (9-10 of piece), X.candacharica (90-115 of piece), A. regeliana (8-9 of piece), D. laeve (15-17 of piece), Ps. trigonochilus (3-4 of piece), Candacharia levanderi (10-14 of piece), Macrochlamys sogdiana (10-12 of piece), Succinea putris (5-6 of piece), Z. nitidus (15-18 of piece). In mountainous areas, C.nitens, C.lubrica, Novosuccinea evoluta, V.costata, Ps.s trigonochilus, P. muscorum, Turanena tenuisira, D. reticulatum, Turcolimax nanus, Z. nitidus are widely distributed throughout the region, relatively high, with an average of 10 to 14 of piece per square meter. Among the stone piles under the bushes and on the stem of various herbaceous plants Ps. albiplicataus (15-17 pieces), Ps. sogdianus (11-14 pieces) species are common. According to the results of the research, the Ferghana valley is a diverse area of species composition of mollusks, distributed in deserts, Adyr and mountain ranges, of which more than 24 species of terrestrial mollusks are considered intermediate hosts, and the degree of density of their population depends on the climatic conditions of the region in which they live.

## CHEMICAL CONTROL OF MELOIDOGYNE INCOGNITA CAUSING ROOT KNOT DISEASE IN CHILLI

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Four commercially available chemical pesticides namely, Actara, Furadan, Ulala and Rugby with six different concretions (50 ppm, 100 ppm, 250 ppm, 500 ppm, 1000 ppm and 2000 ppm) were used to control root knot

infection in chilli plants all the tested pesticides more or less effective to control *M. incognita* and responded positive to chilli plant growth. Although the higher doses of tested pesticides were found more effective for the controlling root knot nematodes such as 0 *M. incognita* has been recorded in roots and soils Furadan (1000 ppm and 2000 ppm) as well as Rugby (2000 ppm) treated chilli plants followed by 0.25 and 0.025 Actara 2000 ppm in roots and soils, and 0.25 in soils in Ulala 2000 ppm as compared to control plants whereas, nematode population was 90.75 in roots and 32.75 in soils. No root knot have been developed on chilli plants those treated with Actara (500 ppm, 1000 ppm and 2000 ppm) Furadan (50 ppm, 100 ppm, 250 ppm, 500 ppm, 1000 ppm and 2000 ppm), Ulala (1000 ppm and 2000 ppm) and Rugby (250 ppm, 500 ppm, 1000 ppm and 2000 ppm) as compared to control plant where 11.25 root knots were recorded. On the other hand, the maximum root length was observed in Furadan 2000 ppm treated plants (18.25 cm) followed by Rugby and Ulala 1000 ppm (9.2 cm) as compared to control (1.3 cm). In term of root weight, the maximum root weight was notified in control plants 5.25 gm. The highest shoot length and shoot weight were recorded in Actara ppm 14. 25 cm and 4.12 gm followed by Ulala 2000 ppm 11.85 and 3.13 gm as compared to control 2.68 cm and 0.1 gm.

## THE EFFECTS OF DIFFERENT PLANT EXTRACTS ON GASTROINTESTINAL PARASITIC NEMATODES

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The goat and sheep breeding are the major parts of the livestock sector in Uzbekistan. The meat and leather products of these animals are considered highly demanded in daily life of the population. The quality of the products decreases due to health conditions of animals affected by various parasitic diseases. Today, there are chemical drugs have been applied to treat the domestic animals from helminthes. However, these drugs do not only affect the helminthes but they also affect the organism of the animal. At the same time, medicinal plants have a complex effect and quickly leave out from the organism. The aim of the research is to identify plants with anthelmintic features and to examine the effects of extracts on the nematodes in the gastrointestinal tract of small animals. To carry out the experiments, there were used animals' parasitic ostertagiine nematodes (Marshallagia, Ostertagia, Tellodorsagia) and extracts produced from local plants. Nematodes were collected from the intestines of sheep in Tashkent region. To store the nematodes in vitro there was used the nutrient medium Leibovitz L-15 and physiological solution. To study the plants with nematodocytic features there were selected 11 different species of widely spread wild and cultivated plants in Uzbekistan: Allium cepa, A. sativum, Tanacektum vulgare, Perovskia angustifoia, Ailanthus altissima, Artemisia leucoids, Chenopodium album, Ferula foetida, F. varia, Matricaria recutita and Elaeagnus orientalis. The experiments were divided into 11 different groups, which were placed in Petri dishes, and there was applied 50 ml of nutrient medium and 50 gastrointestinal nematodes in each group. There was used L-15 nutrient medium and physiological solution as the means of observation. Each dish, used in the experiment, was filled with 0.1, 0.3 and 0.5 ml of the plant extracts mentioned above. Petri dishes were placed in a thermostat with temperature 28-32° C and the viability of nematodes against the effect of drug was recorded in every seven hours for seven days. Each experiment was repeated for three times. The viability of the nematodes examined in a quite condition by observing their twisting movements and by contacting them with drug needle. It was also observed through binoculars. As the results of the research showed, the nematodes in the experiment started to die from day 3. After 7 days in groups I, III, V, VII, IX and X this indicator was at 60-70%, which the research proves that the toxic condition of these plants are moderate. The experimental extracts II, IV, VI, VIII and XI had a strong anthelmintic effect, which accelerated the death of nematodes and gave a high biological effect of 93.3 to 100%. The nematodes, which were in nutrient environment and in the physiological solution survived until the end of the experiment. The results showed that the toxic effects of extracts from different plants in all groups are varying and viability of nematodes species of the genus of Marshallagia, Ostertagia, Tellodorsagia of the parasites of gastrointestinal tract of animals in the nutrient environment is also varied. Basing on the results of the extracts produced from the plants, which contain the high anthelmintic features, the toxic effects of extracts will be experimented on animals in the future researches.

#### ON THE CULTIVATION OF PARASITIC NEMATODES OF RUMINANTS

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The study of the survival of helminthes in vitro, including Marshallagia marshalli (Nematoda: Ostertagiinae) is only important but also one of the problems that needed both theoretical and practical approaches in study. The solution of these problems will allow studying the laws of development, physiology, biochemical and pharmacological aspects of helminthes. In our research we aim to find a nutritious environment that will maintain the long-term viability of nematodes in vitro. To conduct the research experiments, there was used the parasite nematode M. marshalli of small domestic and wild ruminants. These nematodes were collected from the small intestine and abomasum of sheep in Tashkent region, Uzbekistan. To investigate the viability of the M. marshalli nematode, cell cultures the Leibovitz L-15, Medium-199 and the physiological solutions were used. We concentrated on the cell culture Medium-199 as it was reported in some of the literatures that the conducting the experiment in Medium -199 considered effective. The streptomycin (Streptomycin, 5 mg an antibiotic) was used as an antibiotic solution. In the experiments each Petri dishes were filled with 10 mL of nutrient medium-199, 1 mL of antibiotic solution and 50 marshallagia nematodes. Petri dishes with nematodes were placed in a thermostat with a temperature of 28-32° C, and the viability of the nematodes was monitored every 24 hours (see the table). The viability of the nematodes in a quite twist movement was assessed observing visually or by touching the needle. Moreover, sometimes binoculars were also used to observe. The research findings showed that the culture Medium-199 is the best environment for nematodes to survive (20 days). The next culture Leibovitz L-15, where nematodes lived for up to 14 days. In the physiological solution, nematodes survived only few days (9 days). As a conclusion, a favorable nutrient medium for the longer survival of M. marshalli nematode was nutrient medium-199. The obtained data served as the basis to investigate viability of nematodes up imago stages in vitro, although further research is needed.

# HEMATOLOGICAL AND BIOCHEMICAL ALTERATIONS ASSOCIATED WITH ECHINOCOCCOSIS IN DOMESTIC ANIMALS OF MUZAFFARGARH, PAKISTAN

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Hydatidosis is an economically important disease of farm animals and mammals. The present study was carried out to investigate the *Echinococcus granulosis* in sheep, goats, cattle and buffaloes from February 2017 to October 2015 at abattoir of district Muzaffar Garh. A total of 200 animals comprising n=50 sheep, n=50 goats, n=50cows and n=50 buffaloes were examined. ELISa kit of LgG was used for the diagnosis of cysts of Echinococcus. Overall prevalence of hydatidosis was 34.50%. The overall prevalence of hydatid cyst of *E. granulosus* during the study for small ruminants (sheep and goats) were 44.0%, 30.0% respectively, while for large ruminants (cattle and buffaloes) were 46.0% and 18.0% respectively. It indicates that goats and cows serve as an important intermediate host in this region and help in maintaining the cycle. Buffalos were more prevalent of cystic echinococcus as comparison with cows, sheep and goats. In present study, the prevalence in males and females of sheep was 50.0%, 36.36% in buffalos 53.33%, 35.0%, in goats 34.61%, 25.0% and in cows was 25.92%, 8.69% respectively. The prevalence was more in males as compared to the females. The prevalence with respect to age groups in large ruminants (cattle and buffaloes) 36.92% were 45.61% less than small ruminants (sheep and goats). The hematological and biological values of the infected and non-infected ruminants were recorded. The study shows the values of the Neutrophil was lower than the reported means  $36.87 \pm 0.74$  and  $38.72 \pm 0.99$  in normal and infected sheep respectively. The significant decreased was in PCV and platelets in mean

 $36.92 \pm 0.67$ ,  $540 \pm 11.7$  in normal and infected buffalos respectively. The values of leucocytes were lower than that reported mean of  $4.05 \pm 0.72$  and  $7.87 \pm 0.41$  in infected and normal cows respectively. Mean values of RBC, Hb and leucocytes were higher  $31.87 \pm 1.41$ ,  $13.93 \pm 0.76$ ,  $8.06 \pm 0.59$  in infected goats as compared to the values  $12.70 \pm 0.35$ ,  $11.44 \pm 0.10$  and  $7.80 \pm 0.13$  in normal goats respectively. In conclusion, the prevalence of *Echinococcus granulosus* was more in buffaloes, small ruminants and in male. The hematological and biological values of mean and standard error mean were higher in infected animals as compared to the normal animals.

# TRICHURIS TATERI SP.N. (NEMATODA: TRICHURIDAE) FROM TATERA INDICA AND RATTUS RATTUS (RODENTIA: MURIDAE) IN SINDH, PAKISTAN

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A new species Trichuris tateri is described from the small and large intestine of Tatera indica and Rattus rattus respectively. Species of the genus Trichuris Roederer, 1761 are of common occurrence, parasitizing vertebrate host including small mammals. These are reported from various countries including India but none from Pakistan. Present species is reported for the first time from this locality, and *Tatera indica* (Indian gerbil) is a new host record. Species name refers to the host rat. The Tatera indica was trapped from Pipri and Landhi Rice Godowns in suburbs of Karachi while Rattus rattus was trapped from Sujawal Agricultural area in lower Sindh. A total of 256 male and 169 female specimens were recovered from 31 host-rats. Out of 78 rats examined. The specimens were preserved in glycerin + alcohol mixture and were studied in temporary mounts. A new species of the genus Trichuris Roederer, 1761 (Nematoda: Trichuridae) parasitic in the Indian Gerbil Tatera indica and house rat, Rattus rattus (Muridae) from Pakistan is described. The new species *Trichuris tateri* is readily distinguished from *Trichuris musseri* in having a relatively larger body but smaller than Trichuris mallomyos. The resemblance in spicule morphology of the two mentioned species is of special interest because both hosts belong to different tribes and have different habitats and habits. It remains to be revealed whether the resemblance is only homoplasy or actually reflects close phylo-genetic relationship of the parasites. The new species is characterized by the range of spicular length 0.96 - 1.76 (Twenty five specimens studied), upward direction of vulva opening in females, difference in general body dimensions, convoluted tubular testis, vas deferens and seminal vesicle.

## COMPARATIVE EFFICACY OF IVERMECTIN AND ALBENDAZOLE AGAINST ENDO-PARASITES IN PIGEON (COLUMBA LIVIA DOMESTICA)

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The study was conducted to evaluate comparative efficacy of Ivermectin and Albendazole on prevalence of endoparasite in domestic pigeons (*Columba livia domestica*). For that purpose initially 40 domestic pigeons were randomly purchased from different villages and local market of Sakrand, District Shaheed Benazir Abad, Sindh, Pakistan. The pigeons were subjected to coprological examination for the presence of endoparasite. The positive birds were utilized for further experimental protocol. All positive birds with endoparasite weighted and divided into three groups (n=03 in each group) i-e. group A comprised control untreated, group B Ivermectin treated and group C Albendazole treated for evaluation of vivo comparative efficacy of two drugs Ivermectin and Albendazole. The

overall prevalence of positive birds with endoparasites were 22.5%, whereas birds infected with *Capillaria spp.* 10% and *Eimeria spp.* 12.5%. For efficacy of two drugs Ivermectin and Albendazole Fecal Egg Count Rate (FECR) was done on day 1st pre-treatment and 7th and 14th post treatment. The results showed overall efficacy of ivermectin from day 7<sup>th</sup> and 14<sup>th</sup> was recorded 100% against *Capillaria spp.* and 0% against *Eimeria spp.*, whereas albendazole against *Eimeria spp.* was recorded 60% and 0% against *Capillaria spp.* respectively.

# ASCARIDIA GALLI INFECTION INDUCED HISTO-PATHOLOGICAL CHANGES IN BROILER CHICKEN

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In this study, the effect of Ascaridia galli infection on histo-pathological changes in broiler chicken was investigated. The postmortem examinations of broiler experimentally induced A. galli revealed histo-pathological lesions of infected organs viz., lungs and heart. In case of lung, showed chronic bronchitis. Increase in the number of smooth muscle cells. Alveoli were found to be replaced by cystic spaces, and there was cuboidal epithelium. In case of heart revealed atrophy of heart muscles, congestion followed by rupture of blood vessel, fatty degeneration of cardiac muscles, and myocardial necrosis. The fatty infiltration or fatty heart shows an increase in fat cells in the myocardium, were notable observations.

## EPIDEMIOLOGY AND PREVALENCE OF *PEDICULUS HUMANUS CAPITIS* (HEAD LOUSE) IN SCHOOL CHILDREN OF DISTRICT HYDERABAD SINDH PAKISTAN

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Head louse Pediculus humanus capitis are hematophagous obligate ectoparasitic insects, and feed on human blood to survive. Head lice infestation, also known as Pediculosis capitis and nits, is the infection of the head hair and scalp by the head louse (Pediculus humanus capitis). Pediculosis capitis, or head lice, is the most common type of pediculosis, afflicting millions of people annually, mostly school-aged children, in both developing and industrialized nations. Head lice contamination is common worldwide and has been proposed as a major health problem not only in poor countries but also in developed and industrial countries. Head louse infestation considering the importance of students' health, high prevalence of head lice and its complications among the students, and prevalence of this contamination among students of District Hyderabad, we aimed to examine the rate of head lice infestation among the school children of district Hyderabad Sindh in 2018-2019. During the investigation between August 2018 to July 2019 on School children, 2988 children were examined including boys and girls, in this descriptive statically studied students were selected randomly. All the data were collected by questionnaire and also manually examined their heads for checking of nits and adult lice by fine combing, data compile by SPSS. For good statically research students were further divided in three clusters: a) 4-8 age group b) 9-12 age group and c) 13-16 age group. The investigation of school children of district Hyderabad Sindh were conducted during August 2018 to July 2019.All together 2988 children were examined, including boys & girls. Overall 34.29% boys were infested and 66.37% girls were infested. The students were further divided in three clusters i.e. 4-8 age groups, 9-12 age group and 13-16 age groups.4 years to 8 years boys34.36% and girls 59.41%, 9 to 12 years 39.03% boys and 79.90% girls were infested; 13 to 16 years boy's 29.28% and 63.63% girls were infested. Specific training of girls about healthy hair care and prevention of head lice contamination and also its treatment is quite necessary. Monitoring and treating the infested students seem to have a higher priority, too. Because lice contamination in students spreads very quickly and can consequently transmit to their family, it is suggested that the parents be examined and provided with essential training in order to prevent and cure this infestation. Health trainers as member of the school's health group can play an effective role in endorses the health of students. They can provide the students with useful materials like literature in (English, Urdu and in other regional languages) pictures, movies, posters, etc.

### PREVALENCE OF GASTRO-INTESTINAL PARASITES OF HOUSEHOLD DOGS AND RISK PERCEPTION OF THEIR ZOONOSIS IN DISTRICT SWABI KPK PAKISTAN

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Dogs play various roles and their presence within people's houses has increased. In rural and semi-rural and in urban settings dog feaces are not removed from the streets representing an environmental pollution factor. The aim of current study was to evaluate the occurrence of environmental contamination with zoonotic intestinal parasites of five breeds of dogs in District Swabi, Pakistan, with a particular emphasis on GI parasites. We collected 128 dog faecal samples from district Swabi, Pakistan. Coprological analysis performed showed that frequency of positive sample ranged between 80.5 to 92.1% in different breeds. We isolated upto five different parasites in a single sample and detected fourteen helminth and protozoans parasite species. The intestinal helminths detected were Ascaris lumbricoides 28.6% (n=77), Taenia spp 24.5% (n=66), Toxocara canis 19.75 (n=53), Didhylidium caninum 8.55% (n = 23) Toxascaris spp 7.06% (n=19), Ancylostoma caninum 4.83% (n=13), Trichuris vulpis 2.57% (n=8), Echinococcus spp 1.48% (n=4)., Fasciola hepatica 1.11% (n=3), Strongyloides stercoralis 0.74% (n=2), Physaloptera spp 0.37% (n=1). Amongst the protozoans Giardia spp 39.3% (n=13), Coccidia spp 36.3% (n=12), and Isospora spp 24.2% (n=8). Pattern of infection revealed that 19 (14.8%) dogs have single, 42(32.8%) double, 32(25%) triple, and 22 (17.1%) quadruple and 3 (2.34%) pentruple infection. The hunter dogs were highly infected 94.1% (n=17) while the lowest 80.5% (n=21) puppy dogs were recorded. No significant defference was observed in the prevalence of infection with intestinal parasites among these breeds (P >0.05). The result of current study highlight a severe environmental contamination by frequent parasitic stages infectious to humans. There is a higher risk of zoonotic transmission from dogs which indicate an immediate need for the controlling of these parasites and educating the public to take wise action relating to the parasites and pets.

# STUDY ON PREVALENCE AND CONTROL PRACTICES OF GASTRO INTESTINAL HELMINTHES OF SMALL RUMINANTS IN DISTRICT THARPARKAR

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The study on prevalence of gastro-intestinal parasite was conducted in small ruminants of different tehsils/talukas of district Tharparkar. Total 240 faecal sample were collected from sheep (N=120) and goat (N=120) and sample numbers were further divided as 30 sample of sheep and 30 sample of goat from each tehsils/talukas of district. The overall prevalence of gastro-intestinal parasite was 28.75% in small ruminants. Tehsils-wise prevalence of gastrointestinal parasite in goat recorded as 30.00, 30.00, 23.30 and 40.00% in Diplo, Chachro, Mithi and Nangarparkar, respectively. Proportion of gastro-intestinal parasite infection in goat of various talukas was non-significant at P>0.05. Prevalence in sheep was recorded high in Nangarparkar (36.67%) followed by 30.00, 23.33 and 16.67% in Diplo, Chachro and Mithi, respectively. Proportion of gastro-intestinal parasite infection in sheep of various talukas was also non-significant at P>0.05. The gender-wise prevalence of GIT parasite in male was recorded low (24.45%) as compare to female (34.67). Proportion of gander-wise infection in goat was non-significant at

P>0.05. The gender-wise prevalence of gastrointestinal parasite infection in sheep was recorded low in male (21.87%) as compare to female (28.40%). Proportion of gander-wise infection in sheep was non-significant at P>0.05. The age-wise prevalence of GIT parasite infection in goat aging 1-2 years was high (41.82%) and low (20%) was recorded in above 2 year age group of goats. Proportion of age-wise gastrointestinal -parasite infection in goat was non- significant at P>0.05. In case of sheep, the prevalence of GIT parasite infection in aging 1-2 years was high (30%) as compare to above 2 year age group (21%). Proportion of age-wise GIT parasite infection in goat was nonsignificant at P>0.05. Different type of helminths were found in goats during present study, which were *Haemonchus* (35.14%), Trichostrongylus (21.62%), Trichuris (24.32%) and Nematodirus (18.92%). On other hand the helminths of gastrointestinal parasite in sheep were recorded as *Haemonchus* (37.50%), *Trichostrongylus* (18.75%), *Trichuris* (25.00%) and Nematodirus (18.75%). The effect of housing on the rate of gastrointestinal parasite infection in sheep and goat was recorded high (35.82%) in those animals which were reared in open space followed by 20.00 and 16.67% in cottage/chhapra and cemented constructions, respectively. The data regarding to number of small ruminants owner respondent, they use the different source for treatment for their sheep and goats to control the gastrointestinal helminths. The majority of owners used allopathic medication like Ivermactin (53/150; 35.33%), Nilvem (41/150; 27.33%) and Nilzan plus (49/150; 32.66%) while few owners used neem plant as herbal medication (07/150; 4.66%).

## MOLECULAR SURVEY OF PIROPLASMS IN TICK-INFESTED AND TICK-FREE BUFFALOES IN LANDHI CATTLE COLONY KARACHI

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The Piroplasm encompassing two major parasitic diseases; Babesiosis and Theileriosis, have the great veterinary importance both from economic as well as animal health point of view. The study was designed to investigate prevalence of Piroplasms, using molecular and conventional techniques, in buffaloes at Landhi Cattle Colony Karachi Pakistan. The overall prevalence of Piroplasmosis was 15% in examined buffaloes of Landhi Cattle Colony Karachi, Pakistan. Whereas, 18 and 12% of prevalence was found in tick-infested and tick-free buffaloes, respectively through microscopy. The frequency of Piroplasmosis was 60 and 40% in tick-infested and tick-free buffaloes. This difference is considered to be statistically non-significant at (P>0.05). The risk factor was calculated as 1.5, which means the tick-infested buffaloes were 1.5 times more prone to risk of piroplasm as compared to tickfree buffaloes. According to the molecular diagnosis the overall infection of piroplasm was recorded as 23%, whereas 28 and 18% prevalence of piroplasm infection was recorded in tick-infested and tick-free buffaloes, respectively. The frequency of infection was high (60.87) and low (39.13) in tick-infested and tick-free buffaloes, respectively. In case of PCR diagnosis, the risk factor was determined as 1.55 which means that tick-infested buffaloes were 1.55 times more vulnerable to infection as compared to tick-free buffaloes. The prevalence of Theileriosis and Babesiosis was 10 and 8% through microscopy while 16 and 12% through PCR in tick-infested buffaloes, respectively. The frequency of Theileriosis and Babesiosis was 55.56 and 44.45% through microscopy while 55.56 and 44.44% through PCR in tick-infested buffaloes, respectively. This difference is considered to be statistically non-significant at P>0.05 level. The prevalence of Theileriosis and Babesiosis was 8 and 4% through microscopy while 10 and 8% through PCR in tick-free buffaloes, respectively. The frequency of Theileriosis and Babesiosis was 66.67 and 33.33% through microscopy while 55.56 and 44.44% in tick-free buffaloes, respectively. This difference is considered to be statistically non-significant at P<0.05 level. The prevalence percentage was recorded as 8, 6 and 4% through microscopy, while 12, 10 and 6% through PCR, respectively in all examine under study buffaloes for Theileria, Babesia and mixed infection, respectively. The frequency of Piroplasmosis was 44.44, 33.33 and 22.22% in Theileria, Babesia and mixed infection through microscopy, while 42.86, 35.71 and 21.43% through PCR, respectively. This difference is considered to be statistically non-significant at P>0.05 level. The prevalence percentage was recorded as 6, 4 and 2%, respectively in all examined buffaloes at Landhi Cattle Colony Karachi, Pakistan through microscopy, while 8, 6 and 4% by PCR for Babesia, Theileria and mixed infection, respectively. The frequency of Piroplasmosis was 50.00, 33.33 and 16.67% in *Theileria*, *Babesia* and mixed by microscopy, while 44.44, 33.33 and 22.22% by PCR, respectively. This difference is considered to be statistically non-significant at P>0.05 level.

# MORPHOLOGICAL STUDIES ON NATURAL CYSTICERCUS FASCIOLARIS INFECTION IN LIVER OF THE HOUSE MOUSE (MUS MUSCULUS) OF NOSHERO-FEROZE, SINDH, PAKISTAN

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An investigation on the presence of larval cestodes in the House mouse (*Mus musculus*) was carried out in region of Noshero-feroze, Sindh. Morphology of multifocal hepatic cyst found in five of thirty two hosts (15.62%). House mouse were dissected for the presence of hepatic cyst. The creamish to white cysts were present with varying size. Morphological studies of these cysts depicted the presence of metacestodes of *Taenia taeniaeformis* i.e. *Cysticercus fasciolaris* inside them. Scolex of metacestode were mounted carefully and adequate pressure was applied to the cover slip because the hooks to lie flat on glass slide for further study. Meta scolex of specimen were more or less oval in shape, broader than long; T-shaped hooks at rostellum were alternate with large and small hooks in rows; total number of hooks are 38; large hooks are 20 in numbers and small are 18. For morphometric analysis, six variables were studied, number of hooks per rostellum, shape of hooks, total length of large and small hooks and blade length of large and small hooks were considered. Measurement of larger hook vary from 3.70-4.30 in mm and smaller hooks 2.2-2.4 mm. In second row the smaller hooks are placed, these are also variable in length. The smaller hooks vary from 1.8-1.9mm. Length of blade is 1.90mm and length of handle is 1.85mm.

# SERRASENTIS BATHELI N.SP. (RHADINORHYNCHIDAE: SERRASENTINAE) FROM COMMON DOLPHINFISH CORYPHAENA HIPPURUS (PERCIFORMES: CORYPHAEIDAE) OF GWADAR COAST, BALOCHISTAN, PAKISTAN

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The present study is the part of research work on the metazoan parasites of Common Dolphin fish, *Coryphaena hippurus* of Gwadar coast, Balochistan, Pakistan. During present study, a total of 30 hosts were collected from Gwadar coast, Balochistan, Pakistan. Freshly captured Dolphinfishes were transported to the Department of Zoology, University of Sindh, Jamshoro for helminthological investigation. The examination of gut contents and visceral organs revealed the presence of acanthocephalan belonging to genus *Serrasentis* Van Cleave, 1923. The present species differs from its close allies in shape and body size; having 25 longitudinal rows of hooks with 13 hooks in each row of proboscis; distribution of comb-like spines commencing from the lower level of proboscis running ventro-laterally up to the posterior teste, and other characteristics. The new species is named as *Serrasentis batheli*.

## DIVERSITY OF HELMINTH PARASITE COMMON TEAL ANAS CRECCA (ANSERIFORMES: ANATIDAE) MIGRATING TO SINDH PROVINCE, PAKISTAN

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Common teal (*Anas crecca* L.) is a small lduck which breeds widely in Asia from the borders of the Arctic tundra to steppic latitudes and is a winter migrant to Pakistan. They are widespread in Punjab and Sindh and

wherever there is a suitable swampland or water in Balochistan and Khyber Pakhtunkhwa. Common teal are omnivorous, and their diet consists not only of small invertebrates, which they collect with their bill by "da-bbling" under water with their tail up (the characteristic feeding behavior of the sub-family dabbling ducks, the Anatini), but also tadpoles, small fish, or all sorts of plant material which may harbor with the helminthes parasites so the host birds were selected to examine the helminthes parasites. Common teal *Anas crecca* (Anseriformes: Anatidae) werecollected from various water bodies of Sindh Pakistan during December 2019 and January 2020 for the diversity of helmith parasite birds are brought to parasitological laboratory of Zoology Shah Abdul latif University Khairpur her the birds were examine all the birds were effected with the helminthes parasite among them three genera of termatode parasite *Paramonostomum Luhe*, 1909. Echinostoma Rudolphi, 1809 Genus: *Apatemon* szidat, 1928 and a single genus *Polymorphus* Lühe, 1911 of acanthocephalan were observed.

# DAMAGING EFFECTS OF *MELOIDOGYNE INCOGNITA* ON RESISTANT AND SUSCEPTIBLE CUCUMBER CULTIVARS

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In the present study, the damaging effects of six inoculum densities of *Meloidogyne incognita* were compared on a resistant (Long Green) and a highly susceptible (Mirage) cultivar of cucumber. All the inoculum densities of *M. incognita* resulted in significant reductions in growth and yield variables of both the cultivars over their controls. The reductions in resistant cultivar were significantly lower as compared to the highly susceptible cultivar at all inoculum levels. The highest inoculum level caused the maximum reductions in growth and yield variables while the lowest inoculum level resulted in the minimum reductions. The reductions in growth and yield variables increased with an increase in the inoculum density showing a positive direct relationship. On the other hand, the inoculum levels caused an increase in root weight. The higher inoculum levels caused higher increases while at lower inoculum levels, the increases were lower. The increases in root weights were significantly lower in case of resistant cultivar as compared to the highly susceptible one showing a direct relationship between the increase in root weight and inoculum levels. Similarly, significant differences in number of galls and egg masses were noticed between the resistant and highly susceptible cultivar at all inoculum levels. The galls produced on highly susceptible cultivar were significantly higher as compared to the resistant one and direct relationships were observed between inoculum levels and number of galls and egg masses. On the other hand, all the inoculum levels varied significantly regarding reproductive factor on the resistant and highly susceptible cultivar.

# NEOECHINORHYNCHUS DALERI N. SP. (ACANTHOCEPHALA: NEOECHINORHYNCHIDAE) IN THE MARINE FISH MUGIL CEPHALUS (LINNAEUS, 1758) FROM KARACHI COAST, PAKISTAN

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A new acanthocephalan of the genus *Neoechinorhynchus* Hamann, 1892 (Acanthocephala: Neoechinorhynchidae Southwell et Macfie, 1925) parasitic on *Mugil cephalus* Linnaeus, 1758 from Karachi Coast, Pakistan is described. The genus *Neoechinorhynchus* is relatively more common in freshwater fish than in marine environments and some species are known to parasitize turtles. The male and females of this species are less than 7 mm long, proboscis small in size, with the largest hooks being the anterior most 0.098-0.106 in length, lemnisci with 4 nuclei equal in length reaching the anterior testis. Body hypodermal nuclei are 2-3. Central ganglion spherical and located at the base of proboscis receptacle. Lemnisci equal in length reaching anterior testis, oval testes and small bursa. It is closest to

*Neoechinorhynchus dorsovaginatus* but differs in having body size smaller, lemnisci does not overlap anterior testis, lemnisci equal in length, oval testes, cement gland with no obvious nuclei and having smaller bursa.

# HARACTERIZATION OF THE COMPLETE MITOCHONDRIAL GENOME OF CAVISOMA MAGNUM (SOUTHWELL, 1927) (ACANTHOCEPHALA: PALAEACANTHOCEPHALA), FIRST REPRESENTATIVE OF THE FAMILY CAVISOMIDAE, AND ITS PHYLOGENETIC IMPLICATIONS

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The phylum Acanthocephala is a small group of endoparasites occurring in the alimentary canal of all major lineages of vertebrates worldwide. In the present study, the complete mitochondrial (mt) genome of *Cavisoma magnum* (Southwell, 1927) (Palaeacanthocephala: Ehinorhynchida) was determined and annotated for the first time, which represents the first species of the family Cavisomidae with the characterization of the complete mitochondrial genome report. The mt genome of this acanthocephalan is 13,594 bp in length, containing 36 genes plus two noncoding regions. The *trn*V and NCR1 in the mitochondrial genome of *C. magnum* have different positions compared to all the acanthocephalan species available in GenBank. Phylogenetic analyses of amino acid sequences for 12 protein-coding genes using Bayesian inference (BI) supported the class Palaeacanthocephala and its included order Polymorphida to be monophyletic, but rejected the monophyly of the order Echinorhynchida. Our phylogenetic results also challenged the validity of the genus *Sphaerirostris* (Polymorphida: Centrorhynchidae). The novel mt genomic data of *C. magnum* are very useful for understanding the evolutionary history of this group of parasites and establishing a natural classification of Acanthocephala.

## MOLECULAR PHYLOGENETICS AND MITOGENOMICS OF THREE AVIAN DICROCOELIIDS (DIGENEA: DICROCOELIIDAE) AND COMPARISON WITH MAMMALIAN DICROCOELIIDS

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The Dicrocoeliidae are digenetic trematodes mostly parasitic in the bile ducts and gall bladder of various avian and mammalian hosts. Until recently their systematics was based on morphological data only. Due to the high

morphological uniformity across multiple dicrocoeliid taxa and insufficient knowledge of relative systematic value of traditionally used morphological characters, their taxonomy has always been unstable. Therefore, DNA sequence data provide a critical independent source of characters for phylogenetic inference and improvement of the system. We examined the phylogenetic affinities of three avian dicrocoeliids representing genera Brachylecithum, Brachydistomum, and Lyperosomum, using partial sequences of the nuclear large ribosomal subunit (28S) DNA gene. We also sequenced the complete or nearly complete mitogenomes of these three isolates and conducted a comparative mitogenomic analysis with the previously available mitogenomes from three mammalian dicrocoeliids (from 2 different genera) and examined the phylogenetic position of the family Dicrocoeliidae within the order Plagiorchiida based on concatenated nucleotide sequences of all mitochondrial genes (except trnG and trnE). Combined nucleotide diversity, Kimura-2-parameter distance, non-synonymous/synonymous substitutions ratio and average sequence identity analyses consistently demonstrated that cox1, cytb, nad1 and two rRNAs were the most conserved and atp6, nad5, nad3 and nad2 were the most variable genes across dicrocoeliid mitogenomes. Phylogenetic analyses based on mtDNA sequences did not support the close relatedness of the Paragonimidae and Dicrocoeliidae and suggested non-monophyly of the Gorgoderoidea as currently recognized. Our results show that fast-evolving mitochondrial genes atp6, nad5 and nad3 would be better markers than slow-evolving genes cox1 and nad1 for species discrimination and population level studies in the Dicrocoeliidae. Furthermore, the Dicrocoeliidae being outside of the clade containing other xiphidiatan trematodes suggests a need for the re-evaluation of the taxonomic content of the Xiphidiata.

# CHARACTERIZATION OF THE COMPLETE MITOGENOME OF CENTRORHYNCHUS CLITORIDEUS (MEYER, 1931) (PALAEACANTHOCEPHALA: CENTRORHYNCHIDAE), THE LARGEST MITOCHONDRIAL GENOME IN ACANTHOCEPHALA, AND ITS PHYLOGENETIC IMPLICATIONS

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Species of *Centrorhynchus* (Polymorphida: Centrorhynchidae) commonly parasitize various falconiforme and strigiforme birds worldwide. In the present study, the complete mitochondrial genome sequences of *Centrorhynchus clitorideus* was sequenced and annotated for the first time based on specimens collected from the little owl *Athene noctua* (Scopoli) (Strigiformes: Strigidae) in Pakistan. The complete mt genome sequences of *C. clitorideus* is 15,884 bp in length and contained 36 genes [two rRNA genes (*rrn*L and *rrn*S), 22 tRNA genes and 12 protein-coding genes (PCGs) (lacking *atp*8)] and two non-coding regions (NCR1 and NCR2), which represents the largest mt genome of acanthocephalan reported so far. In order to assess the systematic position of *C. clitorideus* and the interrelationship of acanthocephalans from the family Centrorhynchidae and from families in order Polymorphida, the phylogenetic tree was constructed using Bayesian inference (BI) based on amino acid sequences of 12 PCGs. Phylogenetic results supported *C. clitorideus* formed a sister relationship to *C. milvus* in Centrorhynchidae, which has a sister relationship to the representatives of Polymorphidae + Plagiorhynchidae. Moreover, our results also revealed the monophyly of Polymorphida and polyphyly of Echinorhynchidae) was also challenged by the present phylogenetic results, which seems to be a synonym of *Centrorhynchus*.

# A NEW SPECIES OF *NEOECHINORHYNCHUS* (EOACANTHOCEPHALA: NEOECHINORHYNCHIDAE) FROM MARINE FISH (*MUGIL CEPHALUS* LINN., 1758) FROM KARACHI COAST, PAKISTAN

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In a survey of fish parasites of (*Mugil cephalus* Linn., 1758) for acanthocephala, four worms were recovered from a single fish out of 16 examined. On detailed examination it was found that the worms recovered belonged to a new species of the genus *Neoechinorhynchus* Stiles and Hassall, 1905. The previous species of the genus reported are *N. johnii* (Yamaguti, 1939) Bilqees, 1972; *N. karachiensis* Bilqees, 1972; *N. formosanum* (Harada, 1938) Bilqees, 1972; *N. gibsoni* Khan and Bilqees, 1989; *N. nickoli* Khan *et al.*, 1999; *N. longiorchis* Khatoon and Bilqees, 2007; *N. brayi* Bilqees *et al.*, 2011; *N. macrorchis* Shaikh *et al.*, 2011; *N. nawazi* Naqvi *et al.*, 2012. The present specimens have body aspinose, long cylindrical, widest at the end of first half of the body, proboscis round with three rows of six hooks, the largest being of the first row. Neck well developed. Proboscis receptacle single walled, much larger as compared to the proboscis. Lemnisci slightly subequal ending a little anterior to testis. Male reproductive system occupies posterior two third of the body. Testes round to oval, cement gland round, saefftigen's pouch and cement reservoir elongated, bursa small, hypodermic nuclei 5-6, terminal vaginal sphincter and simple uterine bell and eggs oval. Based on these characteristics, the present work describes a new species *N. mohiuddini* from Pakistan. Thus the present work contributes to the knowledge of parasites reported from Pakistan adding a new species. The name *N. mohiuddini* is in honour of Late Dr. Ahmed Mohiuddin, University of Sindh, Jamshoro, Sindh, Pakistan.

## ANTICESTODAL ACTIVITY OF TYPHA ANGUSTATA EXTRACT IN COLUMBIA LIVIA DOMESTICA (DOMESTIC PIGEON) IN DISTRICT DIR LOWER ,KHYBER PAKHTUNKHWA ,PAKISTAN

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Rellitineasis is a helmintic and fatal infection of *Columba livia domestica* (Domestic pigeon) imposing heavy economic losses to human society. Present study was aimed to evaluate the efficacies of crude aqueous extract of *Typha angustata* and sulphadimidine (commercially available drug) for control of rellitineasis. A control based cross-sectional analysis of stool samples were collected from 30 pigeons of 1 year age divided into 3 groups (A, B and C). One was control and other two were drugs exposed groups was carried out (May to August 2019). Two stool samples were collected from each pigeon before diagnosing and after treatment. The samples were fixed in 10% formalin and observed under the light microscope using the methods of direct smear in Lugol's solution, normal saline and flotation techniques. On the basis of drugs accessibility all the Rellitina infected pigeons were divided in to 2- groups. Pigeons in group A were treated with the crude aqueous extract of *Typha angustata* (10 ml per week, progressively increases by 5 ml per week) orally, group B was treated with orally sulphadimidine at a concentration of (10 mg progressively increases by 5 mg per week) respectively. Eggs per gram (EPG) of faeces of infected pigeons were counted in each group performed on Days; 0 (pre-treatment) 7, 14 and 21, 28 (post-treatment). The % efficacies of *Typha angustata* crude aqueous extract and sulphadimidine against rellitineasis infection was reported as 13% and 82% respectively. Present study was concluded that sulphadimidine is the drug of choice for the treatment of rellitineasis in *Columba livia domestica* (domestic pigeons).

## PARASITIC CONTAMINATION OF VEGETABLES COLLECTED FROM DIFFERENT MARKETS OF DISTRICT SWAT KP, PAKISTAN

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The current study was conducted to find out the presence of parasites on fresh vegetables retailed in the different markets of district Swat KP, Pakistan. Total 694 samples of 18 different types of vegetables were collected randomly from three markets of Swat and examined microscopically by using direct smear, sedimentation and floatation techniques. The result showed that 184 samples of vegetables were positive for the presence of parasites. The prevalence rate was 26.51%. Among the detected parasites Ascaris lumbricoides 37.16% was most prevalent followed by Hookworm 21.95%, Entamoeba spp 11.48%, Strongyloides stercoralis 9.79%, Hymenolepis nana 5.74%, Taenia spp 4.05%, Moniezia spp 3.37%, Balantidium spp and Fasciola spp 1.68% each, Trichuris trichuira 1.35% Giardia spp and Sporozoan spp 0.67% each while Schistosoma spp 0.33% was least prevalent. Similarly, Root vegetables 87/200 (43.5%) were most contaminated followed by leaf vegetables 42/162 (25.92%), while fruit vegetables 55/332 (16.56%) were least contaminated. Contamination rate was high in main vegetables market Mingora 28.3% followed by vegetables market Khwaza Khela 26.2%, while the least contamination was recorded in Matta vegetables market. Furthermore, vegetables not washed before display contaminated 37.86% twice as compare to vegetables washed before display 19.19%. From the current study it was concluded that vegetables are potential source for the transmission of parasites.

# GASTRO-INTESTINAL PARASITES IN LARGE RUMINANTS OF LOWER DIR DISTRICT, PAKISTAN

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Gastrointestinal (GI) Parasitic infection is a hot issue for cattle management. There is variation of GI parasites effects in sex, age of cattle, drinking water condition, nutrition, and severity of infection. Prevalence of GI parasites among cattle population in Dir lower has not been published and studied. A total of 40 farm rearing cattle were selected randomly in six tehsil namely Tehsil Adenzi, Tehsil Timergara, Tehsil Balambat, Tehsil Tehsil Munda, Tehsil lalqal, Tehsil Khal. Freshly cattle fecal samples were collected randomly from the selected farms during March 2018 till to December 2018 Out of 314 buffaloes and cattle examined 58.59% (184/314) were positive for cyst/oocyst of one or more species of GI Parasites. The prevalence of parasitic infection was higher in Baffaloe 63.55% (75/118) as compared to Cow 55.61% (109/196) but the difference was not significant (p>0.05) Entamoeba, spp Moniezia spp Haemonchus spp and coccidia spp were found in this study. The non-treated animals indicated the highest percentage of parasitic infection in cow 57.71% (101/175) buffalo 68.13% (62/91).GI parasite prevalence in female animal were higher female cow 62.58% (87/139) female buffalo 77.33% (58/75) as compared to male. But the difference is non-significant (p>0.05) General Linear modelling shown that the treatment of animal was significantly linked with the prevalence of GI parasites. Yearling calves had the highest rate of GI parasitic infection. Future investigation are necessary to evaluate the economic lost which made due to GI parasites.

# ABSTRACTS PRESENTED AT THE FIRST VIRTUAL CONGRESS OF ZOOLOGY

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#### SECTION - IV

#### PARASITOLOGY

# NEW RECORD OF *PROTOSPIRURA SIAMENSIS* RIBAS, 2012 (NEMATODA: SPIRURIDAE) RECOVERED FROM RAT AND MICE OF DISTRICT HYDERABAD, SINDH, PAKISTAN

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In continuation of the ongoing NRPU research project No. 9412 funded by HEC, Islamabad on the Helminth parasite of Rat and Mice. Rodents are the most widely distributed and the largest group of small mammals worldwide and constitute one fourth of the total mammalian species of Pakistan. Rat and Mice are naturally omnivorous scavenger. During the present investigation, total of 51 Rat (28) and Mice (23) were dissected for the presence of helminthic infection. Among nematodes, the most prevalent species was *Protospirura siamensis* Ribas, 2012 reported for the first time from Pakistan. Previously this species is recorded from South- East Asia from the hosts *Bandicota*, *Berylmys, Chiropodomys, Hapalomys, Leopoldamys, Maxomys, Mus, Niviventer* and *Rattus*.

# HYMENOLEPIS DIMINUTA RUDOLPHI, 1819 (HYMENOLEPIDIDAE: CYCLOPHYLLIDEA) OF HOUSE RAT RATTUS RATTUS FROM DISTRICT HYDERABAD, SINDH, PAKISTAN

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In continuation of the ongoing NRPU research project No. 9412 funded by HEC, Islamabad, a total of 72 house rats were examined for the collection of helminths. Out of these, only 31 house rats were infected with 134 specimens of a cestode *H. diminuta* Rudolphi, 1819. On the basis of morphological features like body shape and size, unarmed suckers, segments broader than long, three oval shaped testes separated by ovary, cirrus sac smooth, long and anterior slightly curved, ovary multilobed and round, vitelline gland present above the ovary at middle of the mature proglottids, unilateral pore present and eggs oval or round in shape. The present specimens are identified as *H. diminuta* Rudolphi, 1819. However, present species is recorded for the first time from Hyderabad, hence, making it as new locality.

#### FISH PARASITES IN AQUACULTURE WITH CHANGES IN CLIMATE

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Fish lice are among brachyuran crustaceans, fish lice are the animals reported to parasitize both marine and freshwater fishes. Climatic changes are affecting the attacks and making aquaculture more challenging. *Argulus* spp may be a chief hazard to fish wellbeing, as substantial invasions can be a reason of significant injury and mortality. In addition, fish lice are commonly known to be the reason for other fish infections. Different fish species in different environmental conditions and aquarium were selected to find out the effect of environmental conditions on parasites

and hosts. During different rounds, Argulus spp was poised from the caudal and anal fins of aquarium fishes including goldfish (Carassius auratus). These goldfish may be asymptomatic, and no surplus cases may be celebrated after manual removal of the lice. As soon as any Argulus animals are recognized, control, management and cure may be recommended because contagions can intensify promptly. Currently, there are no FDA-approved medications for the control and handling of this parasite, but numerous compounds and medicines as well as organophosphates and diflubenzuron have been used with achievement. The transmission and isolation of inward bound fish is the best way to avoid an admission of Argulus swarm.

## PREVALENCE OF TICKS AND MITES INFESTATION IN LIVESTOCK AND DRUG'S EFFICACY AGAINST THEM IN DISTRICT DERA ISMAIL KHAN, KPK, PAKISTAN

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The current study aimed to determine the ticks and mites Infestation in livestock and drug efficacy against them in Dera Ismail Khan, Khyber Pakhtunkhwa, Pakistan. In this study, a total of 957 samples in which 375(39.19%) cows and 582 (60.1%) buffalos were examined. Total 794 species of ticks and mites were observed in which ticks were 582 (73.3%) and 212 (26.7%) were mites, which causes Infestation on cow and buffalo bodies. The overall infection percentage of male Sample was 38.42%, while the femaleswere 41.87%. The drug's efficacy is also noted by applying drugs on animal's bodies. Ticks and mites were present more on the udder part, perineum, the lower abdomen, genital areas, and ears of the animal body. In addition, we identify the species (Tropilaelaps spp, Haemaphysalis spp, Amblyomma spp, Mesostigmata spp, and Tropilaelaps spp. It was concluded that the drug trichlorfon 50 ml, when applied on cow and buffalo body surface, has highly effective against ticks and mite's treatment compared to kerosene plus sour oil 100ml.

## EVALUATION OF NEMATICIDAL EFFECTS OF *CANNABIS SATIVA* L. AND *ZANTHOXYLUM ALATUM* ROXB AGAINST ROOT-KNOT NEMATODES, *MELOIDOGYNE INCOGNITA*

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In view of the recently increased interest in developing plant origin nematicides as a surrogate to chemical nematicides, the present study was carried out to assess the nematicidal potential of two antagonistic plants *Cannabis sativa* L. (Cannabaceae) and *Zanthoxylum alatum* Roxb. (Rutaceae) against the most devastating root-knot nematode, *Meloidogyne incognita* responsible for colossal yield losses in cucumber. The leaves of *C. sativa* and *Z. alatum* were incorporated in the soil at the rate of 0, 2, 4, 6, 8, 10 and 20 g per kg of soil. After decomposition, cucumber (cv. Royal Sluis) seeds were sown and inoculated with 2000 s stage juveniles of *M. incognita* ten days after emergence. Data on growth variables and nematode infestations were recorded after six weeks of inoculation. Both the plants significantly reduced nematode infestations and enhanced plant growth criteria compared to the untreated check. The reductions in number of galls, egg masses, nematode fecundity and build up caused by *C. sativa* were significantly higher as compared to *Z. alatum*. Maximum reductions in these variables were recorded with 20 g dosage. The addition of C. sativa and Z. alatum to the soil as organic amendment can work very well as nematicides and can be successfully used for controlling root-knot nematodes replacing traditional chemical treatments and avoiding environmental pollution.

#### INTERACTION BETWEEN MELOIDOGYNE JAVANICA AND RALSTONIA SOLANACEARUM IN CHILI

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The information on the interaction between root-knot nematodes and bacterial wilt is lacking in chili, therefore, in the present study, effects of Meloidogyne javanica and Ralstonia solanacearum were studied on chili singly and in combinations. Significant effects of M. javanica and R. solanacearum and their interaction were observed in case of shoot and root lengths and weights of chili. The mean lengths and weights of shoot and root varied significantly as a result of both the pathogens applied singly and in combinations. The reductions in these parameters were significantly higher when both the pathogens were applied simultaneously as compared to their sole applications. Similarly, at higher inoculum densities, the reductions were the maximum in all these parameters. Significant variations were observed in number of galls and egg masses per root system in response to both the pathogens applied individually and in combinations. Maximum numbers of galls and egg masses were observed where nematode was applied alone. Significant reductions in galls and egg masses were recorded where nematode and the bacterium were applied in combinations. At higher densities of both the pathogens, the reductions in these parameters were the maximum. The first wilt appearance was observed after 8 days in the treatment where both the pathogens were applied at their highest densities. The treatments where bacterium was applied individually at different densities, wilt took maximum days to appear as compared to the treatments where bacterium and nematodes were applied in combinations. Similarly, the incidence of bacterial wilt was lower where the nematode was absent. Minimum wilt incidence was recorded at the lowest density of R. solanacearum. The wilt incidence increased significantly when nematode was inoculated along with the bacterium and maximum incidence was recorded where both the pathogens were applied at their highest densities.

## BIOEFFICACY OF TRICHODERMA SPECIES AGAINST JAVANESE ROOT-KNOT NEMATODE, MELOIDOGYNE JAVANICA, IN GREEN GRAM

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Root-knot nematodes are mainly controlled by using synthetic nematicides, but their excessive use is prohibited due to associated health hazards which demand for suitable alternatives. The overreliance on nematicides can be curtailed by using biological control agents possessing nematicidal or nematostatic properties. Therefore, in the present study, effectiveness of seven indigenous species of *Trichoderma* were tested for their ability to suppress the population of Javanese root-knot nematode, Meloidogyne javanica, and improve growth variables of green gram. All the Trichoderma species resulted in an increase in shoot and root lengths and shoot weight while a decrease was observed in root weight. Maximum increase in shoot length (45.5%) was found in case of T. harzianum followed by T. hamatum and T. viride whereas the increase was the minimum where T. pseudokoningii and T. koningii were applied. Similarly, maximum increase in shoot weight was recorded with T. viride (56.1%) followed by T. harzianum (55%) and the minimum with T. pseudokoningii. As regards root length, it was the maximum in treatments with T. hamatum (46.2%) and T. harzianum (45.1%) and minimum with those where T. koningii and T. pseudokoningii were applied. Contrarily, maximum reduction in root weight was observed in treatments where T. harzianum (37.8%) and T. viride (35.8%) were applied while T. koningii and T. pseudokoningii resulted in minimum decrease. All the Trichoderma species significantly caused reductions in the number of galls and eggs and reproductive factor of the nematode over control. Maximum reduction in numbers of galls and eggs were observed with T. viride (49 and 53%) followed by T. harzianum (46 and 53%) while the minimum reduction was recorded with T. pseudokoningii followed by T. atroviride. Likewise, T. viride caused the maximum reduction in reproductive factor of M. javanica (81%) followed by *T. harzianum* (78%) and *T. asperellum* (75%). On the other hand, the minimum reductions in reproductive factor were observed with *T. pseudokoningii* and *T. koningii*.

## MORPHOLOGY AND ECTOPARASITES OF BROILER AND JUNGLE FOWL CHICKENS (GENUS GALLUS) FROM DISTRICT JAMSHORO AND HYDERABAD, SINDH PAKISTAN

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The comparative input of poultry to total animal protein production in the world is increasing; especially increase in developing countries like Pakistan. Many types and species of ectoparasites (Lice, Mites, Fleas, Ticks etc) and endoparasites (cestodes, nematodes, trematodes etc) are known to infect chickens; which is the major cause of reduction of chickens. This research was based on morphology and ectoparasites of broiler and jungle fowl chicken of (Genus *Gallus*), from two Districts of Sindh which was carried out in the year 2020 to 2021. Total 25 poultry farms were visited and 200 specimens of ectoparasites were collected. While taxonomic work in progress. Prevalence of ectoparasites in the both species will be known after the identification of parasites.

# PARASITOLOGICAL MANIFESTATION OF COCCIDIOSIS IN CAPTIVE BIRDS AND BROILER CHICKENS

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Coccidiosis is an intestinal infection which is commonly known as bloody diarrhea, caused by intracellular protozoan parasite of Eimeria species of Kingdom Protozoa, Phylum Apicomplexa, Class Coccidia, Order Eucoccidioridae, Family Eimeridae, Genus Eimeria. Eimeria species are host specific. coccidiosis is abundently found in captive birds due to contaminated feed and fur. Coccidiosis is sub-clinically found almost in all samples. The current research was aimed to find out the prevalence of coccidiosis infection in Captive Birds and Broiler Chicks in Lahore division. Fecal samples of chicken, pigeons, ducks, peacocks, partridges and pheasants were collected from Lahore Zoo, Tollinton Market, Data Darbar Market, different pluck shops and domesticated birds. 206 Chicken, 122 pigeon, 78 duck, 21 peacock, 102 petridge and 96 pheasant fecal samples were collected during rainy season. 123 chicken (59.70%), 105 pigeon (86.06%), 11 ducks(14.10%), 2 peacock(9.52%), 17 partridges(16.67%) and 19 pheasant(19.79%) samples were positive. The samples were tested by direct smear method and simple flotation method. It was observed that the infection was present sub-clinically in the birds of zoo while the infection was found abundantly in pigeons and layers hens in local markets where the birds were filled in small spaced cages crowdedly. It was also observed that due to lack of sanitary care the feed and water were highly contaminated with feces. The coccidial infection spreads due to overcrowding, contamination, high temperature and humidity. According to our research findings the infection was most abundantly found in Data Darbar and Tollinton Market due to lack of proper health care.

# STUDY ON SEASONAL PREVALENCE OF HELMINTHS IN *CYPRINUS CARPIO* FROM CHILYA FISH HATCHERY, THATTA, SINDH, PAKISTAN

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This study was conducted to assess the prevalence of helminth parasites in *Cyprinus carpio* at Chilya, Fish Hatchery, Thatta. Sindh from February 2019 to January 2020. A total of 107 samples of *Cyprinus carpio* were tested during the research and helminths were found in 17 out of 107. Various helminths groups were identified including trematode and acanthocephalan. Other helminths groups, such as cestodes and nematodes were not noted. These helminths infected the gut of *Cyprinus carpio* including trematode and acanthocephalan. No additional organs were found to be infected with helminths parasites. Helminths prevalence were documented as 41% in September, 40% in August, 36% in October, 33% in June, September (30.76%) and April (25%). During September and October, the helminthes were most abundant, with no infections documented in January, February, March, July, November, or December. Helminths were found in this study throughout specific months of the year.

### **SECTION - V**

# FISHERIES, ECOLOGY, WILDLIFE, FRESHWATER BIOLOGY AND MARINE BIOLOGY

- 1. FRESHWATER BIOLOGY AND FISHERIES
- 2. MARINE BIOLOGY
- 3. PALAEONTOLOGY
- 4. WILDLIFE, DIVERSITY AND CONSERVATION
- **5. BIODIVERSITY**

#### 1. FRESHWATER BIOLOGY AND FISHERIES

### SEASONAL VARIATIONS IN HEAVY METALS TOXICITY AND QUALITY CONTROL ANALYSIS OF SELECTED FISHES FROM RIVER CHENAB

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The present study was conducted from February 2016 to January 2017 to examine the accumulation level of Cadmium, Chromium, Copper and Lead in four fish species (*Labeo calbasu*, *Cirrhinus mrigala*, *Wallago attu* and *Sperata sarwari*) captured from River Chenab, using the technique of Atomic Absorption Spectrometry. The highest significant difference was observed for Cd accumulation in different species as *L.calbasu* (0.596±0.27), followed by *S.sarwari* (0.577±0.21), *W.attu* (0.355±0.06) and *C.mrigala* (0.212±0.04). But there was no significant difference observed for values of Cr, Cu and Pb in *L.calbasu*, *C.mrigala*, *W.attu* and *S.sarwari*. It was observed that significantly highest value of accumulation was of Pb in *W.attu* (1.000±1.70), whereas, the lowest concentration (ppm) of Cd was present in *C.mrigala* (0.212±0.04). The present study revealed that the value of bioaccumulation of selected heavy metals cadmium, chromium, copper and lead were found comparable to the permissible level set by WHO/FAO.

### EFFECTS OF ANTIBIOTICS AND PLANT ADDITIVES (ALLIUM CEPA&ALLIUM SATTIVUM) ON GROWTH PARAMETERS OF LABEO ROHITA

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Due to elevated plea of fish meat to meet nutrient need of humans, use of different supplements (Natural & Synthetic) are in use now a days to enhance growth rate. Fish feed is also trending new technologies To evaluate effects of Phyto-additives (garlic & onion) and Antibiotic (Linco-mycine) on growth, blood parameters and body composition was carried out.80 fingerlings of Labeo rohita were housed in 8 tanks for eight treatments with 2 replicates of each. Treatment was divided into 4 groups depending on their form T1, T2, T3, T4 were fed on powdered feed while T5, T6, T7 and T8were fed on pelleted feed. T1 and T5 were control group with 25% CP level and no additives, T2 and T6 were fed on 25% CP level diet containing 5g kg<sup>-1</sup> garlic and onion each. Fish T3 and T7 were fed on 30% CP containing diet plus 5g kg<sup>-1</sup> diet, T4 and T8 were fed on 35% CP with inclusion of 200mg kg-1 of Linco-mycine. Statistical analysis revealed significant rise in growth and survival in T8, T7 shows significant improvement in comparison to other treatments and control group, but inclusion of Linco-mycine shows significant reduction in RBC's, WBC's. Hb, and Hematocrit but phyto-additives shows significant improvement in blood parameters. Linco-mycine also shows significant drop-in enzymatic activity of amylase & lipase. Biochemical analysis shows significant decrease in protein, fat and phosphorus but significant rise in fiber. It is concluded that Linco-mycine may enhance growth but not safe to use due to its negative effects on hematology and body composition, whereas phyto-additivies also promote growth in comparison to control group so, phyto-additivies are safe to use in aquaculture diet as supplement.

#### AQUACULTURE IN PAKISTAN UNDER CLIMATIC CHANGE CONDITION

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Aquaculture has a limited emission of greenhouse gas as compare to beef, poultry and other activities. It has also a limited impact on deforestation, a limited amount of liquid and solid wastes per kg of meat produced, and a better adaptation to the climate change due to the specific physiology of fishes. In early 1960s, Life Cycle Analysis was developed in order to estimate the ecological influences of a product or provision which can be readily applied to estimate the global warming potential of different types of aquaculture. With Indian major carps, Chinese carps, tilapia and cat fish culture is introduced in Pakistan. Among culture systems, both intensive and semi intensive systems are used. Fish cage culture system is also increasing in Punjab. Life cycle assessment showed global warming potential variations among semi intensive systems.

## GROWTH PERFORMANCE AND SURVIVAL OF GENETICALLY IMPROVED FARM TILAPIA (GIFT) AND *PENAEUS VANNAMEI* (SHRIMP) IN MONO AND POLYCULTURE

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The purpose of the present study was to compare the growth performance of *Penaeus vannamei* and Genetically Improved Farm Tilapia (GIFT) in polyculture system and growth performance of GIFT Tilapia in monoculture system reared during 2016 and 2017 in earthen ponds of Faisalabad Fish Hatchery. It was observed that, GIFT Tilapia cultured individually gained same weight and growth as cultured with shrimp. In 2016, 20,000 shrimp was stocked in monoculture system for growth purpose. Mono cultured shrimp didn't survived and 100% mortality was observed. Polyculture of shrimp with GIFT Tilapia was successfully cultured. During 1st phase, 20,000 post larvae (PLs) of shrimp (*Penaeus vannamei*) were stocked with 5000 tilapia fry and were fed 30% crude protein feed with the intent of residual feed to be consumed by shrimp. Culture duration was six months and average body weight of shrimp harvested remained 20g and Tilapia 490g. This was followed by another trial in 2017, where average harvest weight of shrimps remained 22 g and tilapia 550g in same length of culture period. So, it is concluded that shrimp culture with GIFT tilapia seems beneficial. No extra feed was required for shrimp growth. But overall shrimp growth performance was good and achieved ideal weight for harvesting.

## IMPROVEMENT OF CYPRINUS CARPIO FINGERLINGS PERFORMANCE FED ON TAURINE SUPPLEMENTED OIL SEED MEAL BASED DIET

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Organic acids and their salts could be utilized as acidifiers to improve the performance, wellbeing and resistance status of aquatic living beings. Incorporation of natural acids in fish feed decreases pH in the gut and improves the feed digestion, diminishes the pace of gastric exhausting and improves mineralization. Current study was conducted to evaluate the effects of dietary taurine addition in linseed meal-based diet on growth performance,

nutrient digestibility and mineral retention in Common carp (*Cyprinus carpio*) fingerlings. *C. carpio* fingerlings were fed with graded levels (0, 2, 4, 6, 8 and 10gkg<sup>-1</sup>) of dietary taurine for 70 days. The diets were fed to triplicate groups of fish fingerlings (6.22 g average initial body weight) to apparent satiation twice day and feces were collected. Growth performance and highest survival (98%) rate was significantly improved (P<0.05) by dietary taurine up to optimal level of 4gkg<sup>-1</sup> after which a noticeable decline was observed due to poor retention of various essential nutrients and minerals. Highest digestibility values of crude proteins (73%), crude fat (66%) and gross energy (72%) were observed at this optimum inclusion of dietary taurine in linseed meal-based diet. Likewise better absorption (P<0.05) of majority of minerals (Ca, Cu, Na, P and K) were found in fish fed at the said taurine level. On the basis of these results it was concluded that taurine supplemented linseed meal based diet at the rate of 4 gkg<sup>-1</sup> level is necessary for maximum performance of common carp fingerlings.

# EFFECT OF DENSITY ON GROWTH PERFORMANCE OF SNAKEHEAD FISH CHANNA STRIATA (BLOCH) REARED IN CEMENTED TANKS

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Aim of Present study was to explore the impact of density on survival and growth rate of striped snakehead Channa striata reared in cemented cisterns, Shaheed Benazir Bhutto, University of Veterinary and Animal Sciences, Sakrand Sindh started from February to April 2018. Three different densities (10fish/m², 20 fish/m² and 30fish/m²) were designed to know the suitable density. Results of current research work showed that growth of experimental fish, Channa striata exhibited variation among the experiments. Experiment II (20fish/m²) revealed highest survival rate and growth within the experiments. The average gain in weight of the experimental fish in experiment II was noted higher (16.4 g) in comparison to (11.7 g) and (8.8 g) in treatment I and III respectively. The specific rate of growth and survival was resulted highest in experiment II (0.48 and 100%) in relation to (0.36 and 90%) and lowest (0.31 and 80%) from experiment I and treatment III respectively. Based on the result of present experiment, farmers could be suggested to rear Channa striata at 20/m² density to get higher growth and survival in a short period of time.

# IMPROVEMENT IN NUTRIENT DIGESTIBILITY, MINERAL ABSORPTION AND GROWTH PERFORMANCE OF TILAPIA (*OREOCHROMIS NILOTICUS*) FINGERLINGS FED ON PHYTASE SUPPLEMENTED MORINGA BASED DIETS

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A seventy days experiment was conducted to study the effect of phytase added diets, based on *Moringa oleifera* derived products on growth efficiency of Tilapia (*Oreochromis niloticus*) fingerlings. Tilapia were fed with diet made

up of *M. oleifera* seed and leaf meal mixture. Anti-nutritional factors present in plant derived products, lessens nutrient edibility, utilization of minerals and show negative impact on development. Seven diets were made with addition of PHY (0, 500, 650, 800, 950, 1100 and 1250 FTU per kg). Tilapia were fed at the rate of 4% of live wet weight two times daily and faeces were collected. Based on results, it was noticed that phytase addition indicated huge improvement in development i.e. WG (24g), weight gain percentage (356 %), FCR (1.21), SGR (1.69), nutrient digestibility (CP; 74%, EE; 70%, and GE; 69%) and absorption of mineral (Ca; 72%, Na; 71%, K; 71% and P; 75%) at\_950 FTU kg<sup>-1</sup>. It was additionally noticed, that phytase addition diminished the release of nutrient and minerals through faeces, decreases the water pollution. It was noted that\_phytase addition at level of 950 FTU kg<sup>-1</sup> was useful to formulate cost-effective and eco-friendly fish feed by utilizing Moringa derived products as compared to control and other test diets.

## GROWTH PATTERN AND MORPHOLOGICAL VARIATION OF *LABEO CALBASU* FOUND IN INDUS RIVER, SINDH-PAKISTAN

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The present work reports the length-weight relationships (LWR) and condition factor relationships for Labeo *calbasu* collected from Upstream (Matyari) Kotri barrage at, River Indus, Pakistan, because stock assessment helps the fisheries managers to conserve the commercially important fish. Morphological characters of fish as well as Length-weight relationship are an important tool for fishery management. The results of LWR (W= aL<sup>b</sup>), for *L calbasu*. Representing negative allometric growth pattern. LWRs and condition factor relationships were found significantly correlated. A total of 200 and 190 specimens from upstream and downstream were collected, respectively. The estimated values of length-weight relationship and condition factor were calculated as Kn=39.663 (LeCren), and K=11.915 (Fulton) for upstream and Kn=44.066 and K=13.872 for downstream. Length-weight was found with a strong correlation of n= 2.892, a=0.0235 with r<sup>2</sup>=0.934 for upstream population then the downstream population. The results of this work would be beneficial for sustainable management as well as fishery managers.

# ENVIRONMENTAL IMPACT OF WATER QUALITY ON FISH PRODUCTION IN HALEJI LAKE, DISTRICT THATTA, SINDH, PAKISTAN

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The present studies to investigate the environmental impact of water quality on fish production in Haleji Lake, District Thatta, Sindh, Pakistan. During November 2017 to January 2018 the physicochemical parameter like temperature, pH, dissolved oxygen, Chloride, conductivity, salinity, and total dissolved solids were recorded fortnightly at 8:0AM in each sampling date through -out the study period. In the present study the temperature values were ranged between (20- 23°C with mean 22±1.0), pH (7.3-7.9 with mean 7.62±0.09), DO (4.0 – 5.3 with mean 5.3±0.19), Chloride (0.311-0.375 with mean 0.346±0.028) conductivity (950-1240with mean 1162.5±60.15), salinity (0.3) and total dissolved solid (410-549 with mean 525.7±19.13) from all the sampling sites. In current investigations on the relationship of length and weight of 6 different species were presented, total 140 fish were caught between September 2017 and March 2018 from Haleji Lake District Thatta, Sindh. *Notopterus chitala, Mastacembelus armatus, Notopterus notopterus, Channa punctatus, Aorihthyes aor* and *Oreochromis mossambicus* was enumerated at different length groups. The length weight relationships and correlation of co efficient was analyzed. In the present studies values of b varied from 3.74 *Notopterus chitala*, 2.97 (*Mastacembelus armatus* 2.12 *Notopterus notopterus* 

2.24 Channa punctatus 2.48 Aorichthys aor 3.89 Oreochromis mossambicus. The studies reveled that Oreochromis mossambicus showed better growth and pursued cube law (b=3.89) followed by Notopterus chitala (b=3.74) while Mastacembelus armatus (b=2.97). Aorichthys aor (b=2.48) and Channa punctatus(b=2.24) exhibited closed to ideal. L/W values and coefficient of condition showed ideal growth of six different species from Haleji Lake, District Thatta, Sindh, Pakistan. It is concluded that the different parameters used in the study revealed that the physicochemical parameters in the Haleji Lake is considered to be safe limits (FAO 2012) and good to support the survival and production of aquatic environment especially fish.

#### DAPHNIA'S BREEDING FOR NATURAL FOOD FOR FISH FARMING

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In Uzbekistan, in order to protect the environment in the field of agriculture and fisheries, extensive reforms are underway, with particular attention being paid to the hydrobiological regime of artificial and natural reservoirs, water pollution, the ecology of aquatic organisms and increasing the productivity of reservoirs. In the fauna of Uzbekistan, among zooplankton, more than 20 species of daphnia (Cladocera: Daphnia) are found. Some species among them are distinguished by high biomass growth, adaptation by an external environmental factor, a wide range of vital functions relative to temperature (4-30°C) and resistance to various toxic environments. The studies were conducted in 2019 in Andijan region in a farm specialized in fish farming "Asatilla asl kuli". It was determined by the Daphnia the initial degree of provision of the base with natural food on 31 lakes (with an area of more than 420 hectares). The natural feed base was not recorded at 94.5 % of the total area. For research purposes, over 4 tons of organic manure was introduced on 1.2 hectares of the lake. A pond 10 m long, 5 m wide, 0.8-1.0 m high and 40-50 m<sup>3</sup> in volume was created near the lake for breeding Daphnia. 10 kg of organic manure (cattle) was added to the reservoir per ton of water. After that, they provided water in the amount of 35-40 cm and this amount was saved in the spring and autumn months. In May-August, with a rise in water temperature, the water level was also increased (0.8-10 meters). Using a kapron cone-shaped planktonic net, 200 g of mass were taken from a natural lake and added to a pond where Daphnia was cultivated. Relatively 1 m<sup>3</sup> of water in an amount of 20 g of yeast was added to the walls of ponds where Daphnia was collected. Within 15 days, the productivity of daphnia increased by 50 l of water by ~30-50 g, and in the next breeding cycle by 50-70 g. After that, 3/2 of the water bodies with daphnia were introduced into the lake allocated for research and biomass was measured. The largest amount of biomass was observed in July-August. Along with this, it was determined that in the spring and summer months, one female daphnia develops within 5-8 days, and winter and until half the spring within 40-50 days. As a result of the study, the most favorable time for natural and active parthenogenetic reproduction of Daphnia is the months of March-May and August-October. One daphnia (female) gave 1.4-1.5 kg of product over 36 days (in May-August, one daphnia per during the week increased by 200 units (the number of living individuals)) in the second week by 40,000, and in the fifth week by 1-1.2 million. Organic fertilizers (cattle, horse, and birds) were used to develop daphnia. In conclusion, breeding daphnia in fisheries increases the vitality of fry for which daphnia is the original natural food. This contributes to solving the food problem in Uzbekistan and providing the population with a fish product.

### STUDIES ON THE REARING TECHNIQUES AND BIOLOGY OF CHANNA PUNCTATUS IN CEMENTED PONDS IN SINDH PAKISTAN

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The development of quality fish feed is a key to the success in commercial fish culture. A large number of indigenous raw materials mainly poultry by-product meal, blood meal, various oilcakes, cereal by-products, leaf meals etc. are available within Pakistan. These raw materials can be used in developing supplemental feed for rearing and culture of different fish species. The present study was carried out to investigate the effect of different feeds items and microhabitats on growth and survival of *Channa punctatus* reared in cemented cisterns over a period of three months starting from March to May 2019 and June to August respectively. The experimental fish were obtained from Badin District, Sindh. Three different types of feeds, namely, APC Meal (all parts of chicken meal) feed, Formulated feed, Blood Meal (Treatment I, II and III) having two replicates. The results on the growth performance in relation to different feed items was obtained on all parts of chicken (APC) followed by fishmeal. While lowest growth performance was obtained in the blood meal. While the growth performance in relation to different microhabitats revealed best growth within PVC pipes followed by the mudpots.

# SEASONAL VARIATIONS OF PHYSICOCHEMICAL PARAMETERS IN RELATION TO FISH BIODIVERSITY OF NURRRI LAKE, DISTRICT BADIN, SINDH, PAKISTAN

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Freshwater biodiversity constitutes a vitally important component of the planet, with a species richness that is relatively higher compared to both terrestrial and marine ecosystems. Biodiversity is the term used to describe life on earth — the variety of living things, the places they inhabit and the interactions between them. Nurreri lagoon is situated in Golarhi sub-division of Badin district, 190 kilometers South East of Karachi. This is a natural wetland, featuring a combination of brackish coastal and inland lagoons and barren mudflats on the Northern side. Present study was undertaken to investigate the seasonal Variations of Physicochemical Parameters in relation to Fish Biodiversity from Nurrri Lake, District Badin, Sindh. 372 samples were collected from Nurerri with 2 families having 181 species of *Aulopareia ocellata* and 191 species of *Liza subviridis* and analyzed at different length groups, 2.0 cm each ranged between 1.47 - 35.0 cm were recorded. Among the species *Liza subviridis* was considered as the most abundant and constitutes about followed by *Aulopareia ocellata* of the total catch. The length weight relationship values of the above fish were calculated and observed from the equation that both species were found to be in ideal condition.

### COMPARATIVE STUDY OF OLFACTORY MORPHOMETRIC TRAITS AMONG 11 FISHES BY MULTIVARIATE ANALYSIS

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Olfaction is a highly essential sense in fish and facilitates all the daily life processes such as feeding, locating predator or prey, communal interrelation, reproduction, migration, shoaling etc. In Perciform fish, there are great discrepancies in olfactory organs, related to specific systematic groups and niches. This study will provide information about the differences in olfactory morphometric traits in few perciform fishes belonging to different families including Drepane longimana, D. punctata, Epinephelus diacanthus, Nemipterus japonicus, Pomadasys kaakan, P. maculatus, Rastrelliger kanagurta, Rhabdosargus sarba, Scomberoides commersonnianus, Sillago sihama and Sphyraena obtusata. Out of these, R. kanagurta, S. commersonnianus and S. obtusata are pelagic while remaining are demersal. Now a days, advanced methods are being employed for comparative morphometric investigations such as multivariate analysis. In this study, 15 morphometric traits associated to olfaction were measured in specimens preserved in 10% neutral formalin. The data was log-transformed before the multivariate analysis including Principal Component Analysis, Discriminant Function Analysis and Cluster Analysis. The analyses revealed that although all these species have distinct olfactory structures, S. obtusata is quite dissimilar from rest of the species whereas, there is great similarity exist in the 2 Drepane species while little resemblance in the 2 Pomadasys species examined. However, some other species also show slight similarity with species of a different taxonomic group such as similarity between P. maculatus and N. japonicus as well as between R. sarba and P. maculatus. This study depicts that even fish of different families possess similar olfactory characteristics might be as an adaptive strategy to reside in similar niche or due to the phylogenic closeness among those families.

# EVALUATION OF MORPHOLOGICAL VARIATIONS AND SCALE CHARACTERISTICS OF BLACK POMFRET, PARASTROMATEUS NIGER (BLOCH, 1795) COLLECTED FROM QUETTA FISH MARKET OF BALOCHISTAN, PAKISTAN

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The basic aim of present investigation was to estimate the morphological variation by using morphometric and meristic characteristics amongst the male and female populations of Black Pomfret fish, Parastromateus niger collected from local fish market of Quetta city of Balochistan during the period from June 2018 to July 2019 with size ranged from 12.4 to 33.5cm in Total Length (TL). The variations in detail structural pattern of fish scales collected from eight different body regions were also observe by using Scanning Electron Microscopy (SEM) in the present study. The overall results of this investigation revealed that all selected morphometric characteristics shows strong and significant correlation (r >70; p<0.05) with the total body length (TL), except eye diameter (ED) that indicate a moderate (r = 64.2 %; p<0.05) and significant correlation with whole body growth (TL) for combined sexes, while shows strong correlation (r>70; p<0.05) for females and weak correlation (r<0.50; p<0.05) for males, respectively. On the other hand, the meristic characters showed week and insignificant correlation (r<0.50; p>0.05) with whole body growth (TL) for males, female and combined sexes, respectively. Analysis of 2-sample t-test at 5% significant revealed that significant variations (t-test; p<0.05) were observed in morphological characteristics among the both sexes except eye diameter (ED), snout length (SL) and body depth (DB) and anal-fin ray count (AFC) that showed insignificant variation (t-test; p>0.05); thence reveals the variations among both sexes based on these morphological characteristics. Detail structure of scales also exhibit variation in different body parts of males and females. The shape of the scale may vary from ovoid to oblong. Small sizes of scales were observed in the head and caudal region. Focus was found to be in the middle of the scale in all the scales of male and female except in head region, where it was slightly more towards the anterior portion of scale. Focus was round or oval in shape. Radii were absent. Thus, from the obtained results of scale characteristics, it was concluded that all these scale characters could also be helpful in systematic classification and may consider as a reliable phylogenetic and taxonomic tool.

# A STUDY ON MEASUREMENTS AND COUNTS OF DIFFERENT PARAMETERS OF SCALES OF *PLATYCEPHALUS INDICUS* (LINNAEUS, 1758) FOR FISH IDENTIFICATION

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This study was carried out to find the basic scale characters which are helpful in fish identification. For this purpose, scales of *Platycephalus indicus* (Linnaeus, 1758) were collected from six body regions. Scales were cleaned with NaOH and dehydrated with different grads of alcohol (30%, 50%, 70%, and 90%). Slides were prepared for study under the microscope. The results of the study show that the dorsal side of the fish was covered with ctenoid scales and the ventral side was covered with cycloid scales. Radii were observed at the anterior part of the scale. No radii were investigated at lateral or posterior margins of the scale. Focus was present near to the posterior part of the scale. The shape of focus was observed to be circular, oval and pear shape. In general, the size of the scales was ranging between 2.0mm-4.2mm in TLS and 1.0mm-2.9mm in WDS. The radius (Rs) was ranging from 0.3mm to 1.8mm. The number of radii was observed between 8-15. The ctenii were present only in the scales of dorsal side of fish body, ranging 12-35 in HRS and 2-9 in VRS. This study provides basic knowledge about structure of scales of *P. indicus* and about the measurements and counts of different parameters of scales, which could be supportive in fish identification.

# STUDIES ON MORPHOMETRIC CHARACTERISTICS, GROWTH, MORTALITY AND STOCK EVALUATION OF INDIAN MACKEREL, *RASTRELLIGER KANAGURTA* (CUVIER, 1817) FROM THE BALUCHISTAN COAST OF PAKISTAN, NORTH ARABIAN SEA

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Based on the study of 344 specimens collected monthly from three main fish landing harbors of Baluchistan, Sonmiani, Gadani and Kund Malir, for the period of two years, from January 2007 to December 2008, fifteen morphometric characters, growth, mortality and yield per recruit of Indian mackerel Rastrelliger kanagurta, were studied. The morphometric lengths (total length, fork length, standard length, head length, pre-dorsal fin length, preorbital length, eye diameter, lower-jaw length, pre-anal fin length, head depth, pectoral fin length, pelvic fin length, greatest depth and pre-pelvic fin length and pre-pectoral fin length) were recorded and the descriptive statistical parameters and correlation coefficient (r) were analysis with independent variable (total length) and dependent variable (other morphometric parameters). The result shows that morphometric parameters are highly correlated to total length. The length weight relationship data of the present study shows negative allometric growth (b = 1.719 to 2.376). The von Bertalanffy growth parameters for combined sexes were  $K = 0.46 \text{ year}^{-1}$ , 1.60 year and 0.62 year. asymptomatic length  $L\infty = 24.73$  cm, 27.83 cm and 23.63 cm and  $t_0 = -2.83$  year, -0.60 year and -2.07 year at Sonmiani, Gadani and Kund Malir respectively. The total mortality coefficient Z was estimated as 2.26 year-1, 3.31 year-1 and 2.14 year<sup>-1</sup>and the naturalmortality coefficient M was 1.13 year<sup>-1</sup>, 2.48 year<sup>-1</sup> and 1.39 year<sup>-1</sup> at Sonmiani, Gadani and Kund Malir respectively. In the present study the calculation indicated an average  $E_{\text{max}}$  to be 0.367± 0.016 against the present average exploitation rate (E) of  $0.366 \pm 0.124$  thus indicating only limited opportunity is existing for production increase from the present grounds along Baluchistan coast, Pakistan. In conclusion the population dynamics and fishery attributes of all important fishes in the Baluchistan coast of Pakistan, North Arabian Sea should be studied for build-up effectual managing strategy.

# FEEDING BEHAVIOUR OF CATFISH, OMPOK PABDA FROM INDUS RIVER NEAR JAMSHORO, SINDH

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Ompok pabda is a freshwater catfish, commonly found in rivers, canals, streams, lakes, ponds, and in undated fields (Mirza 1982). Ompok pabda is medium-sized belongs to family Siluridae of order Siluriformes is native to Pakistan, India, Bangladesh and Myanmar (Banik and Malla (2011). It is delicious, well preferred and high priced due to relatively few bones (Sarkaret al., 2005). The knowledge on feeding biology and behavior is the key for selection of cultural species for stocking purpose. The availability of natural food items in the environment vary with the season, temperature and time of the day (Narejo et al 2016). There is no published information is available on any aspect of the biology of this commercially important catfish from Pakistan. So, that the present study was designed to supply such information and that will serve as a basis for subsequent studies on food feeding behavior of this important freshwater catfish from Pakistan. Few isolated studies carried out by Banik and Malla (2011) and Baniket al (2011) are available. The results of the present studies have practical values and would be useful in future for the development of culture techniques of this species in ponds. Ompok pabda is suitable candidate for the artificial culture in future, so knowledge of various aspects of biology is considered as pre requisite.

Feeding habit To investigate the food and feeding habits of Ompok pabda was enumerate from the guts content which were collected from river Indus near Jamshoro, Sindh Pakistan during three months from March to May 2017. Total 200 specimens ranged from 4.0 -30.0 cm and 6.1-60.5 g in length and weight respectively were procured from the catch of local fishermen. The collected specimens were brought to laboratory preserved in 5% formalin. Collected samples was analyzed in virtue of total length and weight by use of scale (measuring board) and balance (digital balance) and kept in vials for subsequent studies and classified as empty, full and half. Sample gut preserved in formaldehyde were cut up and examined by use of counting chamber following the standard methods for quantitative analysis of plankton sort and recognized on specie level.

The gut contents from the anterior portion of the gut that is stomach were carefully washed into a Petri dish. Dissected and observed under a light microscope (Olympus, model B-2000) by using Sedgwick-Rafter counting Cell. Analysis was followed by the method of Ivlev (1961). Ivlev's index; Electivity (E) was calculated according to following formulae:

$$r1 - P1 E = ---- r1 + P1$$

Whereas, relative contents of items represented as r1, items present in the environment donated (+1) positive value (-1) while negative value resulted as avoidance of food substances. Several workers like Hobson, 1974; Narejo et al 2016; Tesfahun 2018 studied gut contents by using volumetric frequency of gut matters. Below equation was applied for assessment

$$I1 = V1.01$$
  $\sum V1.01$ 

V1 is the volume and % of occurrence I = is the preponderance index of particular items of food respectively. The percentage volume of gut contents was determined by the eye estimation method (Pillay, 1952). The percentage occurrence of all different food items were determined by summarized up to the total occurrence of all items from which the percentage occurrence of each items was calculated (Hynes, 1950). Feeding rate of the fish was determined by calculating the gut content and weight of gut (Gastro somatic index).

The fish sample comprised of a total 200 specimens ranged from 4.0 -30.0 cm and 6.1-60.5 g in length and weight respectively. The food of Ompok pabda was determined from the gut analysis and revealed that the feeding behavior of Ompok pabda was found to be carnivorous with higher feeding preferences for insect larvae (60.43%), followed by zooplanktons (23.582) and 3rd preferred food was debris (11.93). It was noted during the present course of investigation that the small fish (4.0-10.0 cm) preferred zooplankton, protozoa and plant debris while medium sized fish (11.0-16.0 cm) showed preference to insect larvae, worms and detritus and large fish (17.0-30.0 cm)

exhibited insect larvae, zooplankton, detritus and debris. It was concluded that feed preference of Ompok pabda in the present study was carnivorous in feeding habits with soft invertebrates, the percentage of different food items from the gut of Ompok pabda from River Indus. The gape of mouth was measured in relation to total length of Ompok pabda from river Indus and found positively co- related with the total length, that indicated that the mouth gape is increases with the increase in total length of fish

It was concluded that the food of Ompok pabda contained insect larvae, zooplankton, detritus and debris. It indicated that the catfish, Ompok pabda from river Indus is mainly carnivorous in feeding habit and change their feed preference according to size.

### LENGTH-WEIGHT RELATIONSHIP AND RELATIVE CONDITION FACTOR OF HETEROPNEUSTES FOSSILIS (BLOCH) FROM LAKE MANCHAR

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The present study relates the length-weight relationship, relative condition factor of fish species *Heteropneustes fossilis* (Bloch) from Lake Manchar. The fish is separated into male, female and pooled to compute length-weight relationship and relative condition factor separately. The correlation coefficient 'r' exhibits high degree of relationship of growth performance of length-weight relationship in both the sexes and pooled. The analysis of coefficients of regression calculated methods given by LeCren (1951) of *H. fossilis* varied between 7. 38-31.9 cm resulted into equation given Log w = -0.12+2.43 (For male), Log w = -0.16+2.58 (For female) and Log w = -0.53+2.45 (For pooled). It might be observed that the calculations that the values of b were found to be alike and less than 3 (good). The b value observed close to be Ideal growth in female and pooled, while in case of male it indicated as satisfactory growth of Singhee, *H. fossilis* from Lake Manchar. The index of plumpness or wellbeing (Kn) of Singhee, *H. fossilis* from Lake Manchar District Jamshoro, were calculated separately for either sexes and pooled. The mean plumpness values 1.0,0. 93, 0.99 in case male, females and pooled respectively. The mean plumpness values in males were found with lower values throughout the study period in compression with females. Finally it was found that the length weight relationship and Kn of Singhee, *H. fossilis* from Manchar Lake District Jamshoro indicated satisfactory growth (positive allometric growth) for male, female and pooled respectively.

# INDIVIDUAL AND SYNERGISTIC EFFECT OF COPPER NANOPARTICLE AND VITAMIN E SUPPLEMENTATION ON GROWTH, OXIDATIVE STRESS AND FILLET QUALITY OF JUVENILE OREOCHROMIS NILOTICUS

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The Feeding experiment was conducted with juvenile *Oreochromis niloticus* to assess the effect of copper nanoparticle and vitamin E supplementation on growth, oxidative stress and fillet quality. Fish (average initial weight  $11.44\pm0.02$  g) were randomly distributed in glass aquaria (80 L water holding capacity L 45 x H 45 x W 30) at the rate15 fish/aquaria. Four isoproteic (350 g/kg) and isolipidic (75 g/kg) diets were designed named as, T1 (Cu0+VE0 mg/kg), T2 (Cu2+VE0 mg/kg), T3 (Cu0+VE500 mg/kg), T4 (Cu2+VE500 mg/kg). Triplicates were assigned for each treatment for the course of three months applying completely randomized design (CRD). At the termination of experiment, growth was evaluated by weighing each treatment individually. Growth in terms of final weight (g), average weight gain (g) and weight gain % was significantly increased in fish fed with T4 (p< 0.05). Similarly, improved FCR was also observed in the same group, whereas highest survival rate was observed in groups fed with T3 and T4. Liver and gills catalase (CAT) activity (mU/mg protein) and peroxidase (PoX) activity (mU/mg protein) was improved with dietary supplementation of vitamin E (T3) whereas, improved antioxidant enzyme activities were

observed in T4. The whole body unsaturated fatty acids (USFAs) content in *O. niloticus* were significantly increased in group fed with diet T3, whereas muscle amino acid content was not significantly different among all dietary treatments. Hence, the results indicated that supplementation of tilapia diets with copper nanoparticle and vitamin E could reduce oxidative stress and improve the growth and health profile.

# GROWTH AND IMMUNE RESPONSE OF *LABEO ROHITA* AND *OREOCHROMIS NILOTICUS*UNDER THE INFLUENCE OF 2% *NIGELLA SATIVA* SUPPLEMENTED FEED

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Black seed (Nigella sativa) is widely used in aquaculture feed and stimulates the immune response and growth performance of fish. An experimental study was carried out for the assessment of growth and immune response of Oreochromis niloticus and Labeo rohita under the influence of 2% Black seed (Nigella sativa) supplemented diet. The proposed project was carried out in Fish Nutrition Laboratory, Department of Zoology, Wildlife and Fisheries, University of Agriculture Faisalabad. Duration of the trial was eight weeks. Two aquaria, each having the capacity of eighty liters of water, labelled as  $T_0$  and  $T_1$  were used for stocking ten fingerlings of each specie. The experiment was carried out in triplicate under each treatment group. Fish were fed a diet having 40% crude protein at the rate of 6% of body weight daily. Fish feed supplemented with 0 and 2 percent of black seed was given to T<sub>0</sub> (control) T<sub>1</sub> (treatment), respectively. Growth performance and physico-chemical parameters such as temperature, pH, total alkalinity, dissolved oxygen and total hardness were noted weekly. Weight gain by rohu in T<sub>0</sub> and T<sub>1</sub> was 6.32 (g) and 4.96 (g) respectively. Nile tilapia's weight gain in  $T_0$  was 16.78 (g) and in  $T_1$  was 25.76 (g). When compared to the control group, statistically non-significant difference was observed in weight gain of both Labeo rohita and Oreochromis niloticus under treatment T<sub>1</sub>. Other growth parameters calculated for T<sub>0</sub> and T<sub>1</sub> were as following: gain in length of rohu 1.28 (cm) and 1.36 (cm), tilapia 2.16 (cm) and 3.24 (cm), FCR of rohu 0.29±0.107 and 1.2±1.542, tilapia 0.33±0.346 and 0.12±0.025, SGR of rohu 0.97+0.394 and 0.32+0.095, tilapia 1.1+0.221, 2.7 +1.203 respectively. Statistical analysis had shown that FCR and SGR of rohu and tilapia in T<sub>1</sub> were significantly different than that of T<sub>0</sub>. Blood samples of the fish from T<sub>0</sub> and T<sub>1</sub> were also assessed for white blood cell count ( $161.67\pm1.33$ ,  $198.00\pm2.19$   $10^3$ /µL in rohu) ( $139.00\pm1.00$ ,  $169.00\pm1.20$ 10<sup>3</sup>/µL in tilapia), lysozyme activity (40.79, 49.66 ug/mL rohu) (17.83, 30.33 ug/mL tilapia) and immunoglobulins (14.58±0.75, 23.67±0.70 ug/mL rohu), (15.73±0.79, 26.13±0.80 ug/mL tilapia) at the end of experiment. In comparison to the control group, immunological parameters of black seed treated group were significantly increased.

# HEAVY METALS (ZN, NI AND CU) POLLUTION IN WATER, PLANKTONS AND FISH FROM BALOKI HEADWORK TO SIDHNAI BARRAGE, RIVER RAVI, PAKISTAN

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The toxicity of heavy metals viz. Zn, Cu and Ni in water, planktons and fish had studied from Baloki headworks to Sidhnai barrage, river Ravi for six months. All samples were collected from three main sampling site, Baloki headworks, Kamalia-Chichawatni bridge and Sidhnai barrage. Samples of water, planktons and fish collected by using the bottles, planktons net and fishing net, respectively. Pollution of metals in planktons measured on dry weight basis. In all samples, concentrations of heavy metals measured with Atomic Absorption Spectrophotometer and then processed statistically through ANOVA, regression and correlation. Planktons showed maximum pollution than fish and water. Pollution of metals in the water, planktons and fish measured in following order as zinc > nickel > copper. The concentration of metals in samples from all studied stations measured in following order as Kamalia-Chichawatni bridge > Baloki headworks > Sidhnai barrage. In organs of fish, pollution of metals followed the order

as liver > kidney > gills > muscles. The accumulation of metals in fish were above the permissible values. The consumption of fish from the studied locations may pose dangerous health effects to the consumers.

# POTENTIAL USE OF SALINITY TO ENHANCE THE GROWTH OF GIFT TILAPIA IN AQUACULTURE – A SUITABLE CANDIDATE FOR FARMING AT HIGHLY SALINE WATERS OF PAKISTAN

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Competition between agriculture and aquaculture for land and water resources has rendered brackish water as an alternative mean for fish farming. This study aimed to observe the growth performance of Genetically Improved Farmed Tilapia (GIFT) at two cities, Shahpur and Faisalabad at different salinity levels while keeping temperature variations in view. GIFT fry weighing 0.5g was initially stocked in nursery ponds at each site. Feeding was done regularly and growth was monitored fortnightly from June to November, 2016. Monitoring of salinity and other water quality parameters was carried out during the entire study period. Shahpur had comparatively higher salinity (3.00-4.01 ppt) than Faisalabad (2.00-3.00 ppt) and promising results for growth were observed for Tilapia at Shahpur attaining 532g with standard length of 21.9cm. High salinity levels were observed during warmer months and significantly positive correlation was observed for salinity, temperature, total dissolved solids and electrical conductivity. The effect of salinity on fish growth was highly significant (p<0.05) and highest growth rate was observed at salinity levels between 3 and 4 ppt. It could, therefore, be concluded that GIFT tilapia is a remarkable nominee for farming at higher salinity levels compared to other culturable fish varieties which are unable to survive at high salinity and pH. This study would help in creating awareness about using saline water for GIFT farming to meet the dietary needs of people of Pakistan.

# STUDIES ON GROWTH AND SURVIVAL OF *LATES CALCARIFER* REARED WITH *OREOCHROMS MOSSAMBICUS* AND *PUNTIUS GONIONOTUS* UNDER POLY-CULTURE SYSTEM IN FISH FARM OF DISTRICT HYDERABAD.

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The present study was carried out to investigate the effect of Growth and survival of *Lates calcarifer* reared with *Oreochroms mossambicus* and *Puntius gonionotus* under poly-culture system reared in ponds over a period of three months starting from June to August 2019. Three different types of feeds, namely, Treatment I with Live feed, Treatment II with chopped fish flesh and Treatment III with formulated feed having two replicates. The physico-chemical parameters were recorded on fortnightly basis throughout the study period. It was observed that no significant variation were observed in different water quality parameters throughout study period, the temperature values were ranged between 21.40-23.87, pH 8.16-8.37, salinity 0.38–0.43, DO 3.26-3.33, total dissolved solid 470.66-489.66 and conductivity 1171-1181.

# INVESTIGATION OF THE EFFECT OF SELECTED NATURAL (BLACK CUMIN) AND SYNTHETIC PRESERVATIVE (SODIUM ACETATE) ON PROTEIN AND FAT CONTENT OF *L. ROHITA*.

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Fish foods have recently received more attention from consumers due to their positive benefits on human health and nutrition. Due to their high nutritional value, their consumption has risen substantially over the past few decades. The rohu (Labeo rohita) is important aquatic food item also commercial fish species of Pakistan. The aim of this study was to investigate the effect of selected natural (black cumin) and synthetic preservative (Sodium acetate) on the protein and fat content of L. rohita. Natural preservative black cumin seed has some biological activities such as antioxidant, anti-inflammatory, anticancer and antimicrobial. Synthetic preservative such as sodium acetate efficient against spoilage of microorganisms and extended the shelf life. Total 96 sample of species were purchased washed and degutted. Out of 96, 6 samples will be freshly analysed and remaining used as control and experimental group. Natural preservative black cumin and sodium acetate (3g/l, 5g/l and 7g/l) used to enhance the shelf life of fish. From the first day sample were analyzed at every 15 days interval up to 45 days to measure the protein, lipid, moisture and dry matter. Protein and fat contents were determined by kjeldahl method and Soxhlet extractor respectively. In L. rohita highest protein content % observed in control group (24.791±1.263), black cumin seed oil (28.437±1.546), sodium acetate at concentration of 3g/l (26.916±2.162), at concentration of 5g/l (25.520±2.525) and at 7g/l concentration (24.971±1.263) was observed on the day of 15. On the day of 45, minimum value of protein contents % in control group (20.125±1.515), black cumin seed oil (24.791±1.263), sodium acetate at concentration of 3g/1 (23.333±1.262), at concentration of 5g/1 (21.145±1.263) and at concentration 7g/1 (19.687±2.187) noted. Highest fat content % observed in control group (11.67±0.235), black cumin seed oil (10.83 $\pm$ 0.288), sodium acetate at concentration of 3g/1 (10.66 $\pm$ 0.288), at concentration of 5g/1 (10.5 $\pm$ 0.5) and at 7g/1 concentration (9.66±0.577) was observed on the day of 15. On the day of 45, minimum value of protein contents % in control group (7.83±0.288), black cumin seed oil (8.33±0.577), sodium acetate at concentration of 3g/l  $(8.16\pm0.763)$ , at concentration of 5g/l (7.83±0.763) and at concentration 7g/l (7.66±0.763) noted. The results with control group, black cumin seed oil and sodium acetate at concentration 3g/l, 5g/l and 7g/l showed the highly significant relationship of protein and fat contents with the increasing number of day's observation. The value of protein and fat decreased with increasing number of days. This research will help people to preserve fish food by using black cumin and sodium acetate as safe preservative to increase the shelf life of food and maintain the quality for longer time.

# EFFECT OF CINNAMON AND GARLIC ON PROTEIN AND FAT CONTENTS OF ROHU (*LABEO ROHITA*)

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Fish is an important source of food and it fulfills the demand of human nutritional standards due to the presence of key nutrients. It is a cheap source of animal protein and fish oil is an important source of omega-3-polyunsaturated fatty acids, minerals and vitamins. Shortly after the capture, fish begins to deteriorate. Three major mechanisms contributing in spoilage of fish are microbial activity, enzymatic action and lipid oxidation. Due to consumer's awareness of harmful effects of chemical preservatives, extensive researches have been made on use of natural preservatives with antimicrobial and antioxidant properties. This research work was conducted to determine the effect of garlic and cinnamon oil on protein and fat contents of Rohu (*Labeorohita*). In experimental group, samples of fish were preserved in garlic and cinnamon oil and in control group, no preservative was used. All

samples were freezed at 4 ±1 °C and analyzed at interval of 15, 30and 45 days. Protein and fat contents were determined by kjeldahl method and Soxhlet extractor respectively. Garlic and cinnamon treated sample had higher protein and fat content (%) than control group. Maximum protein content for control (24.791 %) garlic oil (28.437%)) and cinnamon oil (29.166 %) was recorded on 15th day of storage. The minimum protein content was recorded on 45th day of storage in control (20.125 %), garlic oil (24.791 %) and cinnamon oil (25.687%). Similar results were observed for fat content. Highest fat content for control (10.500 %), garlic oil (11.166 %) and cinnamon oil (11.333 %) was calculated on the 15th day. Minimum fat content was noted on the day 45 in control (7.833 %), garlic oil (10.00 %) and cinnamon oil (10.160 %). The results with control group, garlic and cinnamon oil displayed the highly significant relationship of protein and fat content with the increasing number of days of storage. Result of this study also revealed that protein and fat contents in both control and experimental groups decreased with increased storage period. From the results, it is concluded that use of garlic and cinnamon extract can represent an alternative way to inhibit the microbial growth and oxidative damage in fish meat and extend shelf life of fish.

# COMPARATIVE STUDY OF ROHU (*LABEO ROHITA*) FRESHWATER FISH IN ROHRI CANAL AND COMMERCIAL PONDS AT KHAIRPUR, SINDH, PAKISTAN

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Freshwater carp farming is the major aquaculture activity in three of the provinces of Pakistan namely (Punjab, Sindh and KPK).Good and adequate nutrition that plays a very important role in the controlling of mental, physical and intellectual qualities in our body. Fish is the only medium which can be served for many purposes such as to reduce blood lipid level, particularly serum triglycerides. In addition, fish is good source for human nutrition due to their therapeutic role in reducing certain cardiovascular disorders. The present comparative study shows that two habitats namely ponds and freshwater (Rohri canal) of Labeo rohita (Rohu) have different growth rate in terms of age, weight and length. This study demonstrates that water physico-chemical factors extensively influence on the growth of fish, therefore the monitoring/exchange of water is vital for optimum fish product. Various locations-based investigations were carried out for estimation of rate of length and weight. It was found that the fish weight is low in the month of February 2018. The obtained results in terms of length and weight for various months including Luqman Khairpur: L=18, W=1235 (Feb) L=18, W= 1316(June), Pir-jo-goth L=18, W=1245 (February), L=18, W=1282, (May).Rohri canal L=18, W=1372 (February), L=18=1398 (June). The physico-chemical parameters of water such as pH, temperature, turbidity, DO, Tds, salinity and conductivity were measured and in the month of June the value of these water parameters were higher and lower results and it is reason that Fish growth is significant in the month of June, 2018. As Pakistan has different climatic changes owing to its four seasons in a year that has great impact on aquatic life. The total number of 150 Rohu (Labeo rohita) fishes were observed from Luqman, Khairpur, and Pir-jo-Goth and Rohri canal in subjective period from February 2018 to June 2018. A significant growth in the freshwater fish strengthen the claim for harvesting Fish carp into the economy.

## WATER QUALITY PARAMETERS AND ICHTHYOFAUNAL DIVERSITY OF RIVER KABUL AT CHARSADDA

### Majid Khan

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A study on the fishes of River Kabul at District Charsadda, Khyber Pakhtoonkhwa was conducted from March 2017 to September 2017. A total of 16 fish species were identified belonging to 4 Orders (Cypriniformes, Siluriformes, Channiforms and Mastacembeliformes) and 8 families (Cyprinidae, Nemachilidea, Sisoridea, Siluridae, Bagridea, Channidae, Mastacembeldae and Schilibeidae). Family Cyprinidae was the richest family of the present survey represented by 5 species; viz Carassius auratus, Tor macroplepis, Barilius modestus, Barilius vagra, Rasbora

daniconius, and the four families consist on two species i.e. Sisoridea, Siluridae and Channidae. Family Sisoridea consist on Glypthothorix punjabensis and Gagata cenia. Family Siluridae was also denoted by two species i.e. Ompak pabda and Wallago Attu. Family Channidae is also consist on two species i.e. Channa punctatus and Channa striata and Family Bagridea is also representing by two species i.e. Mystus bleekri and Rita Rita. All other families were represented by a single species i.e., Mastacembelus armatus (Mastacembeldae) Clupisoma naziri (Schilibeidae) and Acanthocobitis botia (Nemachilidea) Throughout the study period, Clupisoma naziri was the most dominant and abundant genus of the fish followed by Glypthothrix punjabensis, Gagata cenia, Barilius modestus, Barilius vagra, Tor macroplepis, Rasbora daniconius, Carassius auratus, Wallago attu, Ompak pabda, Mastacembelus armatus Acanthocobitis botia Channa punctatus, Mystus bleekri, . In the present survey, many species such as Barilius modestus, Barilius vagra, Carassius auratus, Gagata cenia, Glypthothorix punjabensis, Channa punctata, Tor macroplepis, and Mastacembelus armatus etc. Were common with adjoining rivers like Khiali and Jhindi. Majority of fish spices collected during the survey were edible and play a key role in local economy these includes, Carassius auratus, Tor macroplepis, Carassius auratus, Ompak pabda, Wallago attu, Clupisoma naziri, Canna punctata, and Mastacembelus armatus. Along with fish fauna identification some important water parameters are also measured in the presence of available laboratory material i.e. Alkalinity. PH, TSS, TDS, Water temperature, Air temperature, which are the key indicators of status of fresh water indicating the suitability of water for fish life. The mean value for these that were noted during this study are; Air temperature 26.42 C°, Water temperature is 22.54 C°, velocity of water is 0.35 m/sec, Ph is 7.31, Total suspended solids 125.1mg/l, Total dissolved solids is 115.75 mg/l, Alkalinity 253.33 mg/l, Water depth is 16 feet. The aforementioned values are all the mean values of every parameter measured during the present study period which indicate that River Kabul is suitable for Fish life but furthermore study is required to explore the Fish fauna and to takes some emergency steps to save the important Fish fauna in the precious River Kabul.

## HISTOPATHOLOGICAL AND HEMATOLOGICAL ALTERATIONS CAUSED BY SALT STRESS IN CTENOPHARYNGODON IDELLA

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Our current studies were based upon the freshwater fish grass carp (*Ctenopharyngodon idella*). We investigated the different Hematological and Histological parameters of the grass carp in this experiment. In our trial we exposed the fish to the different concentrations of salt (Sodium chloride) viz., 0, 2, 4, 6 and 8 g/L water. We kept the fish in the above different saline conditions for four weeks. After four weeks we took blood from the caudal vein of the fish and observed different parameters of Hematology viz., WBC count, RBC count, HGB Conc. HCT%, MCV, MCH, MCHC, Platelets count and percentage of Lymphocyte. We observed the significant rise (P≤0.05) in WBC count, RBC count, HGB conc. HCT% and platelets count with the rise of salt conc. while, there were no significant variations shown in the remaining blood parameters. In case of histology we observed three organs of the fish viz., Gills, Kidney and Heart. We observed the proliferation of Gill chloride cells, Gill Hyperplasia, fusion of secondary lamella, removal of the epithelial membrane, degeneration of secondary lamella and mucosal membranes in gills exposed to high salt concentrations. In the kidney we found swelling of glomeruli, clustering of glomeruli, increased Bowman's spaced, increased tubular spaces and tubular cell necrosis. Whereas when we observed the conditions of heart tissue, we didn't find significant variations except increased spacing between the cells of cardiac tissues, degeneration and scattering of ventricular cells.

## MOVING TOWARDS MODERN AND LATEST TECHNOLOGIES IN AQUACULTURE INDUSTRY FOR SUSTAINABLE FISH FARMING PRACTICES

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For meeting the growing desire for food future world's appetite for fish, the improved performance and propagation of aquaculture industry leading in producing more farmed fish per unit of land and water, is highly

recommended. This can be achieved through the adoption of modern and latest technologies / techniques while leaving behind the classical conventional methods. This is surely a very broad spectrum which is the basic potential in way of contribution towards sustainable fish farming systems. It can be accomplished not only through the development, dissemination and application of new biological, chemical and mechanical techniques at the existing and newly developed fish farms but is also dependent upon the knowledge, information, education, training, skills and management of fish farmers and fisheries personnel carrying out all the activities at their on-farm-sites. Through meeting the challenges for facilitation of the right incentive framework and provision of technologies with reference to the specific circumstances, the overall objectives can be achieved leading to the environmental benefits while creating and meeting the sustainable future food demands and sidewise strengthening the domestic or global economy of the region.

## 2. MARINE BIOLOGY

# ASSESSMENT OF A SUBTIDAL MACROBENTHIC COMMUNITY AROUND K2/K3 NUCLEAR POWER PLANTS IN KARACHI, (NORTHERN ARABIAN SEA)

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The sub- tidal macrobenthic faunal survey was conducted in 2015 to study the occurrence of the benthic fauna in the vicinity of the Nuclear Power Plants in Karachi (KANUPP) K2 / K3 outfalls around a radius of 5 km. Subtidal samples were collected from under water rocks through SCUBA diving by a private vessel from different stations in the vicinity of KANUPP, Karachi under the project "Pre-operational biological studies around proposed outfall of K-2/K-3, northern Arabian Sea". The subtidal rocks of the study area have diversified assemblages of macrobenthic fauna which mainly belongs to different phyla such as Cnidarian, Annelida, Arthropoda and Echinoderms. The most dominant organisms were found to be the mollusks, specially *Turitella* and *Epitonium* sp. Other benthic fauna included Polychaetes, mostly represented by *Neries* whereas in crustaceans, amphipods and barnacles were in abundance. Echinoderm was represented only by a brittle star. The hard corals belonging to the genus *Favites* and *Goniopora* sp. were also found during the study. It was further observed that Coral covers, gorgonian density and other fauna were found relatively lower in stations nearest to the outfall, whereas the off shore submerged rocks were preferably rich in corals and gorgonian density.

# COMMUNITY STRUCTURE AND BIODIVERSITY OF MARINE MACRO FOULING ORGANISMS IN THE COASTAL WATERS OF KARACHI, PAKISTAN

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The species composition and diversity of marine macro fouling assemblages in the coastal waters of Karachi, Pakistan were analyzed. Fouling assemblages were obtained from test panels immersed in Korangi Fish Harbour for a period of about 30 days, from December 2016 to November 2017. Some samples of fouling assemblages were also obtained from navigational buoys, boat hull, and oil jetty pillars in Manora Channel, intermittently from October 2017 to May 2018. A total of 67 taxa were recognized, 48 of them up to species level. Polychaete was the most abundant group in terms of number of taxa (23), followed by Arthropoda (20) and Mollusca (11). Species richness was greater in test panels and navigational buoys (41 taxa each) followed by jetty pillars (23) and boat hull (11). Diversity indices calculated for the four communities include Shannon-Weiner (H'), Simpson reciprocal (1/D), Margalef (M) and Pielou (J). Stress-predictability modelling shows that the buoys community was the most stable and favourable. Sorensen similarity coefficient and Bray-Curtis index were highest between buoys and jetty pillars communities and lowest between test panels and boat hull communities.

# ZOOPLANKTON COMPOSITION, DIVERSITY AND ABUNDANCE IN GADANI (BALUCHISTAN COAST)

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Zooplankton are minute aquatic animals that are weak swimmers and drift in water column of ocean, seas or fresh water bodies to move great distance. The species diversity and community structure of the zooplankton are

necessary to assess the potential fishery resources. Due to their small size, short life span, species diversity and high tolerance to the stress zooplankton are used as the indicator species for the physical, chemical and biological processes in marine environment. Triplicate samples were collected from ship breaking area in Gadani each month using zooplankton net, abundance and diversity of zooplankton were recorded employing standard methods. The zooplankton abundance and diversity shows variations with different seasons and varied from station to station. The peak abundance is in South West Monsoon than in Spring inter Monsoon, Autumn Inter Monsoon and North East Monsoon at coastal waters of Gadani. Abundance of zooplankton varied from 176-12631 individuals/m³. The peak zooplankton abundance occurred in April and the lowest abundance was recorded in December. Copepod dominate the zooplankton group, highest percentage was recorded of Calanoid 18.80% than Cyclopoid 14.78% and Herpecticoid 11.44%. The marine fishes graze on the zooplankton and abundance of zooplankton determines strength of commercial important fish species. The information obtained by the present study may be utilized by government and non-government institutions involved in the fishery industry.

# THE PREVALENCE OF THE PEA CRAB, NEPINNOTHERES VILOSULLUS, AND ITS IMPACT ON THE DIFFERENT SPECIES OF CLAMS AND COCKLES IN PAKISTANI WATERS (BALOCHISTAN)

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First report regarding the association between the bivalves and Crustacean decapods Pea crab, *Nepinnotheres villosulus* was detected from six species of clams, *Meritrix casta* var, *ovum, Marcia marmorata, Amiantis. umbonella, Gastrana multangula, Protapes cor* and *Tellinimactra angulata* and in one species of blood cockle, *Anadara antiquata* collected from Sonmiani during May-04 to April-05. The highest prevalence of pea crabs were recorded in *Meritrix casta* var. *ovum* and the lowest was found in *Protapes cor*. Out of 482 specimens of blood cockle *A. antiquata* examined, pea crab were found in 15 cockles (3.11% of the total sample) of which eleven (11) were females and four (4) males whereas infested specimens of clams amounted 49.60 % of clams while remaining 50.40 % of clam were infested simultaneously by two pea crabs species. The rate of overall percentage of females (55.32) infested was slightly greater than the males. The mean size of the male and female pea crab was 8.60±2.60 and 5.30±1.20 mm, respectively. The ovigerous females constituted 62.82% of the total female pea crabs examined during the entire study period. Except for, May, June-04, January and April-05, the ovigerous pea crabs were found in remaining eight months of the sampling period, constituting 45.45 to 75.00% of the total females. The GI of infested clams was low compared to uninfested clams.

# SOME UNDOCUMENTED DECAPODANS FROM THE INDIAN OCEAN IN THE GALATHEID COLLECTION OF IIOE (1963-64) BY THE SHIP ANTON BRUNN

### Quddusi B. Kazmi\* and Feroz A. Siddiqui

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'During 1963-64 Anton Bruun collected 535 specimens of galatheids were sent to the University of Karachi through the courtesy of the Smithsonian Institution for report writing. When the galatheid collection was being returned to the Smithsonian Institution, the senior author noticed several non galatheid decapods mixed with galatheids. They were carideans, porcellanids, juvenile majid crab, juvenile hermit crab and a carapace of penaeid shrimp, all were separated from the main collection and identified. The species were assigned as *Glyphocrangon mabahissae* Calman and *Pontonides unciger* Calman in Caridea while *Petrolisthes militaris* (Heller), *Lissoporcellana demani* Dong and Li (dealt separately), *Lissoporcellana quadrilobata* (Miers) and *Lissoporcellana* sp. (near *L.nitida* 

(Haswell) and two unidentified *Pisidia* (one close to *P. delagaoe* ) in Anomura. All are described and illustrated in this paper.

# BIOLOGY AND MIGRATION: A CASE STUDY OF FRESHWATER PRAWN IN ISLAND AND CONTINENTAL RIVERS.

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Life history migration is an important component of many aquatic animals to complete their life cycle. Migration is a regular and rhythmic movements of animals in terms of season, direction and life stage and migratory animals inhabit in two distinct and well-separated habitats. Those animals which migrate between two different aquatic environments (marine and freshwater) are termed as diadromous. Amphidromy is a common form of diadromy mostly evident in freshwater prawns and fishes. This is a gametic form of migration where animal need marine environment for completion of their larval development. Amphidromy in freshwater prawns have two patterns depend on riverine length

- 1. Ovigerous females spawn in upstream and larvae drift down to the estuarine area: In small length rivers female spawn upstream, small rivers guarantee the lesser reasons of mortality of larvae during downstream drift, including lesser velocities and fewer predators as compare to large rivers.
- 2. Ovigerous females migrates down to river mouth for spawning: Mostly downstream migration of ovigerous female is observed in larger rivers to avoid larval mortality.

Amphidromous prawn has adaptations in reproductive biology to coup up with mortality during downstream larval drift and upstream migration of juveniles. Due to construction of barrages and climate change Indus River is facing water scarcity specially in lower reaches which results in restriction of movements of migratory animals including *Tenualosa illisha* (Palla) and freshwater prawns. In Pakistan River Indus is the life line for human population, agriculture and aquatic life. Indus River is the home to many freshwater prawn species including most important aquaculture and commercial species i.e. *Macrobrachium malcolmsonii* (monsoon river prawn) and Macrobrachium *rosenbergii* (giant river prawn) during a survey of Indus river which was carried out over seven years only four specimen of *M. rosenbergii* were observed. However, *M. malcolmsonii* was found in large number except the months of lower flows in Indus River. An altered flow of Indus Rivers has adversely effected the populations of freshwater prawns.

# COMPARATIVE ACCOUNT OF ESSENTIAL AND NONESSENTIAL METAL CONCENTRATIONS IN FIVE MARINE CRAB SPECIES FROM MANGROVE AREA OF KORANGI CREEK, KARACHI, PAKISTAN

### Asmat Saleem Siddiqui and Noor Us Saher

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Mangrove environment and their allied fauna progressively influenced and endangered worldwide because of unsustainable humanoid activities in the Anthropocene Era. During the past few decades, heavy metal contamination exaggerated along the estuarine and coastal environment of Pakistan. Korangi Creek is one of the areas blessed by the mangrove environment in Pakistan and affected by Malir River runoff and airborne pollution, as a result, the risk of contamination boosted in the food chain. Therefore, the current study aimed to investigate the metal levels in the mangrove sediments and their allied fauna. The concentrations of essential (Zn and Cu) and nonessential (Co, Cd and Pb) metals were analyzed in the sediments and muscles of five brachyuran crab species (Austruca iranica, A. sindensis, A. urvellie, Macropthalmus depressus and Metaplex indica) from the mangrove habitat of Korangi Creek, Karachi,

Pakistan. No significant variation (p > 0.05) found in the accumulation of essential metals, however, the nonessential metals showed a significant difference (p < 0.05) in the muscles of all five studied species of crabs. Nonessential metals presented significant correlation (p < 0.05) among themselves indicated that they accumulated within the crab tissues through similar sources i.e. anthropogenic one. All selected crab species showed a sediment-biota accumulation factor greater than one for all metals, except Co, which specified the high rate of metal accumulation referencing the surroundings. In this study, the mangrove habitat and resident crab species revealed the contamination and the risk of non essential as well as essential metals and expecting for sustainable management and conservative strategies from the stakeholders.

# REDESCRIPTION AND PHYLOGENY OF THE MANGROVE CRAB *ILYOGRAPSUS RHIZIPHORAE* BARNARD, 1955 (CRUSTACEA: DECAPODA: OCYPODIDAE) FROM THE PAKISTAN COAST

### Uroj Aziz<sup>1</sup>, Noor Us Saher<sup>2</sup> and Mustafa Kamal<sup>3</sup>

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Mangrove intertidal areas provide a diverse habitat for the variety of semi terrestrial brachyuran crabs from tropical to subtropical regions of the world. From the coast of Pakistan, the genus *Ilyograpsus* has been represented by a single species which was previously misidentified as *I. paludicola*. The current study based on morphological, morphometrics and molecular analysis, to understand the taxonomic position of *Ilyograpsid* crab from the coast. This study, based on morphological as well as morphometric characteristics suggested that instead of *I. paludicola*, the type species; *I. rhizophorae* is distributed along the coast of Pakistan, which is redescribed. *I. rhizophorae* can be distinguished from *I. paludicola*, by key morphological characters, including proportion of carapace length to breadth, cheliped, merus and gonopod 1 of male crab specimens. The genetic diversity and phylogenetic position of *I. rhiziphorae* were inferred from analysis of 16S mitochondrial DNA using Genbank sequences of Macrophthalmid, Ocypodid and Grapsoid crab species. Maximum-likelihood tree (ML) and Maximum parsimony analysis (MP) showed that *I. rhizophorae* were closer to Macrophthalmid crab species as compared to Grapsoid and Ocypodid crabs.

## MARINE ENVIRONMENT COVERAGE IN THE MASS MEDIA: INSIGHTS FROM NEWSPAPERS IN PAKISTAN

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Science coverage in the print Mass media distressing decline in during current, past, whereas the science journalists paid a heavy price to austerity measures, being frequently among the first to be laid off. Those left in place now dispose of much reduced means for in-depth investigations of what are usually complex issues. As a consequence, a various number of newspapers and magazines have significantly reduced, if not exclusively eradicated, their science pages. Marine environmental news is assumed to be a less commonly reported item in the Mass media. Media has a deleterious effect, so there is a need to find a role and impact of Mass media in the field of Marine environment. Although some media platforms are ignoring marine related news and coverages nevertheless there is a channel through that media founds the impact of the message which communicated to their target audience. The current study investigates Marine environment related news coverage by the Pakistan print news houses and quantified its patterns by reviewing 600 news items across three national newspapers in English languages (Dawn Newspaper, The News Newspaper and The Express Tribune Newspaper). The present study finds its role and coverage effect by the content analysis and would identify that media message transformation is productive or not?

So research will investigate the ratio of information productivity as well, it's a long term relationship between the fields and media so this research is focusing on the marine news the research results will provide more debt on the role and impact of news in this field.

# DISCRIMINANT ANALYSIS OF TWO SPECIES OF MANTIS SHRIMPS *ORATOSQUILLA NEPA* AND *O. INTERRUPTA* FROM THE COASTAL WATERS OF KARACHI, PAKISTAN

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The morphology of Mantis shrimps, *Oratosquilla nepa* (Latreille, 1828) and *Oratosquilla interrupta* (Kemp, 1911) of family Squillidae are exhibiting a significant degree of overlap, therefore the discriminant morphological variance among the two species of *O. nepa* and *O. interrupta* was determined. In this study, the morphometric characteristics examined in samples obtained from the Karachi fish harbor during Dec 2018 to Dec 2019. Morphometric characters (Total length, carapace length, carapaces width, abdominal length, thoracic length, thoracic somite 5, thoracic somite 6, claw length, merus lengths, propodus length, dactylus length and telson-length) transformed and subjected to discriminant analysis that depends on grouping model, number of discriminant functions and revealed the total variance, morphological variance among populations. Specimens belonging to *O. nepa* (N=50) and *O. interrupta* w (N=50) were compared using multivariate statistical analyses of morphometric characters. A stepwise discriminant analysis run in two variants and based on thirteen variables: The success rate of classifying the groups according to the discriminant analysis of morphometric characters, overall, 88% of the observations were placed into the correct group. *O. nepa* had the highest proportion of correct placement, with 92% of the observations correctly placed. *O. interrupta* had the lowest proportion as compare to *O. nepa* and the correct placement, with 21 of 25 observations, or 84%, correctly classified. It is determined that there is a variation between *O. nepa* and *O. interrupta* according to the morphometric characters.

# LIPID CONSTITUENTS FROM RANCID LIVER OIL OF DEAD RHINCODON TYPUS (WHALE SHARK)

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Whale shark *Rhincodon typus* is the biggest shark and biggest extant fish in the world. It is the only member of family Rhincontidae. These highly migratory species with cosmopolitan distribution are found in all tropical and temperate seas. These are currently classified as vulnerable to extinction. Ecological, biochemical and dietary preferences of whale shark are reported through lipid analysis and identification of fatty acids. Analysis of liver oil from a stranded, dead whale shark exploiting GC-MS, GC-FID, and RI has resulted in identification of 62 constituents. 24 were found present in the preferred prey of whale shark. In contrast to 19 SFA, 24 MUFA, and 17 PUFA, reported in fresh liver oil, only 14 SFA, 10 MUFA, and 6 PUFA, (82.85%) were identified in liver oil. Rest have undergone degradation/oxidation. The trends and possible routes of the degradation/oxidation of LCFA, MUFA, and PUFA has been discussed and 33 constituents were justified originating from rancidity, included 21 FAld (7.46%), 2 FAlc (0.59%), 4 ketones (4.19%), and short chain FA. Short chain fatty acids are also found reported as anabolic and catabolic products. 9 FA are being reported in this study for the first time with 5 miscellaneous constituents (2.11%).

# GONADOSOMATIC INDEX, GONADAL DEVELOPMENT AND FEUNDITY OF BLUE SWIMMING CRAB, PORTUNUS PELAGICUS (LINNAEUS, 1758) FROM BALOCHISTAN COAST, PAKISTAN

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This paper deals Gonadosomatic Index, gonadal development and fecundity of the blue swimmer crab, *Portunus pelagicus* (Linnaeus, 1758) from Dam Sonmiani Balochistan coast. For the study of GSI values, total of 110 male crabs were dissected, GSI ranged from 0.70 to 6.20 having a mean of  $3.80 \pm 1.08$  S.D. In female, GSI values were ranged from 0.71 to 13.33% having a mean of  $5.46 \pm 3.25$  S.D. Three stages of male testis development and four stages of ovarian development were observed throughout the study period. Fecundity of *P. pelagicus* was calculated in 65 berried females of 3 different egg stages. 28 berried females of first stage, 13 crabs of second stage, 24 berried female of third stage were calculated for the fecundity. The numbers of eggs were found from 86392-1555820 in various sizes of Short Carapace Width from 70-138 mm. The average fecundity was 27 523773  $\pm$  279204 (S.D) for a berried crab with a mean short carapace width of  $98.53\pm18.05$ . Some smaller crabs were often found carrying more eggs than bigger crabs. The total egg mass weight ranged from 7.5-115 g (mean  $29.53\pm21.30$  SD). Relationships between SCW/ Fecundity, SCW/Egg Mass weight and Total Body Weight/ Fecundity were also calculated.

### A STUDY OF FISH FAUNA OF TANDA DAM KOHAT, KHYBER PAKHTUNKHWA

### Zaigham Hasan and Riaz

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Fish fauna of Tanda Dam Kohat was studied from January to April 2015. During this study, collection was made from different sites of the dam. A total of 308 fishes were collected which comprised of 2 species. The species were identified and found belonging to 6 Orders, 7 families and 17 genera. Order Cypriniformes was represented by 2 families viz family Cyprinidae and Nemacheilidae. Family Cyprinidae was the most abundant family with 15 species viz Labeo rohita, Labeo dyocheillus pakistanicus, Catla catla, Cirrhinus mrigala, Ctenopharyngodon idella, Hyphophthalmichthys molitrix, Cyprinus carpio, Puntius sophore, Puntius ticto, Puntius conchonius, Barilius pakistanicus, Barilius vagra, Tor putitora, Crossocheilus diplocheilus and Aspidoparia morar while Family Nemacheilidae was represented by only one species, Schistura prashari. The rest of the fish species namely Chanda nama, Channa punctatus, Xenentodon cancila, Ompok pabda and Mastacembelus armatus belonging to families viz Chandidae, Chanidae, Belonidae, Siluridae and Mastacembelidae respectively were also reported. During the study period it was also detected that some species were present in large number while some species were present in lesser number. Fish species like Hyphophthalmichthys molitrix, Cyprinus carpio, Labeo rohita, Aspidoparia morar, Xenentodon cancila, Mastacembelus armatus, and Crossocheilus diplocheilus were most common and species like Barilius pakistanicus, Barilius vagra, Tor putitora, Schistura prashari, Chanda nama, Puntius ticto, and Puntius conchonius were less common. During the study it was also found that the economically important stocked species such as Hyphophthalmichthys molitrix, Cyprinus carpio, and Labeo rohita grow very well in Tanda Dam while Catla catla, Cirrhinus mrigala, and Ctenopharyngodon idella were not very successful and showed very poor growth. Further research is required especially on this aspect.

### 3. PALAEONTOLOGY

# GIRAFFID FOSSILS FROM EARLY-MIDDLE MIOCENE LOCALITIES OF PAKISTAN

### Kiran Aftab1\*, Muhammad Akbar Khan2, Zaheer Ahmed3 and Muhammad Shadab1

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The deposits of fossils in the Siwalik Hills of Pakistan are the primary way to understand the diversity and advancement of mammalian evolution. New giraffid fossils have been recovered from Early-Middle Miocene localities of Pakistan. These localities are present in Kamlial and Chinji Formation of Lower Siwalik with estimated age 18.3–11.2 Ma. To collect ancestral forms of mammalian fossils, special attention has been given to the outcrops of Kamlial Formation. The material comprises mandible and maxillary fragments, and isolated upper and lower dentitions. The studied specimens compared with the relevant samples housed in different Paleontological Museums in Pakistan and other foreign countries. Early-Middle Miocene giraffid completely disappeared around 10 Ma from the Subcontinent before the onset of the Dhok Pathan Formation. The presence of *Giraffokeryx punjabiensis* all over the Chinji Formation shows its widespread distribution in the Subcontinent. The Early-Middle Miocene giraffids preferred to inhabit wooded mean forested areas of the Siwalik Group.

# MIDDLE MIOCENE MAMMALS OF CHABBAR SYEDAN IN JHELUM, PAKISTAN: ENAMEL HYPOPLASIA

### Muhammad Khalil Nawaz\*, Muhammad Akbar Khan, Sayyed Ghyour Abbas, Muhammad Asim and Muhammad Akhtar

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Enamel Hypoplasia is the reduction of tooth enamel resulting from interruptions in the enamel deposition by ameloblasts for the period of tooth growth. Overall 135 teeth specimens were analyzed, out of which 13 were found with Enamel Hypoplasia (EH). These fossil teeth have a chronological range of 14.3-11.2 Ma and belong to Lower Siwaliks. The current EH analysis was performed to find out Enamel Hypoplasia occurrence in relation to changing ecological conditions. Enamel Hypoplasia occurrence in mammalian fauna of Chabbar Syedan signifies that there were numerous vegetational and climatic changes during the middle Miocene of Lower Siwaliks.

# DESCRIPTION OF NEWLY RECOVERED REMAINS OF *KOBUS PORRECTICORNIS* FROM LATE PLIOCENE SIWALIKS OF PAKISTAN

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Some new remains of *Kobus porrecticornis* have been discovered form a new locality, Dillo Mora, Tatrot Formation of the Siwaliks, Pakistan. The recovered fossils date back to late Pliocene Siwaliks of Pakistan. (3.6-2.58)

Ma). The sample comprises of upper molars and premolars of *Kobus porrecticornis*. The molars show prominent basal pillar, deep central fossettes with complex spurs projecting anterioposteriorly, prominent anteriotransverse goat fold and strong median ribs between the styles. The premolars are comparatively smaller sized than molars. The lobes are compressed posteriorly and have prominent transverse flange. When compared to previously described fossils of *Kobus porrecticornis* from similar strata of the Siwaliks, considerable similarities in dental characters are present which are diagnostic to this species. The present samples strengthens the already existing but scanty data of *Kobus porrecticornis*. *Kobus porrecticornis* is considered as primitive Siwalik Reduncine which mostly inhabited the mosaic of grassland and woodland of late Pliocene time span along with other grazing families like Bovidae, Elephantidae, Equidae and Cervidae.

# SIAMOTRAGULUS FROM THE CHINJI FORMATION OF THE CHABBAR SYEDAN, JHELUM, PUNJAB, PAKISTAN

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Chabbar Syedan is a small village situated at the base of the Bakrala ridge, Jhelum, Punjab, Pakistan. The outcrops of this village represent the Chinji Formation by lithology and faunal elements. Field campaigns from 2016 to 2019 in these outcrops resulted in the collection of some good specimens including the artiodactyls and perissodactyls. Fossil remains of family Tragulidae are also present. Though small in number but indicate the presence of genus *Siamotragulus*. *Siamotragulus* dental remains have not been described from the Siwaliks. Two specimens are extremely small in size and these may represent a new species. A detailed analysis of these specimens in progress.

## 4. WILDLIFE, DIVERSITY AND CONSERVATION

# MORPHOLOGICAL STUDY OF NUMIDA MELEAGRIS (GUINEA FOWL OR TURKEY BIRD), FAMILY NUMIDIDAE: ORDER GALLIFORMES, IN SINDH, PAKISTAN

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Birds are highly diverse species that are randomly distributed in different regions of world. Numida meleagris (Guinea fowl or Turkey bird) is one of the valuable species that are endemic to Africa and rank among the oldest of the gallinaceous birds, though they are reared as pet in Pakistan. Present study was carried out to record the morphological characteristics of N. meleagris and for this purpose, field surveys were conducted in Larkana, Kambar Shahdad Kot, Sukkur, Dadu and Jamshoro districts of Sindh province. Present study was focused determination of parameters such as body color, structure and color of beak, color of plumages (dorsal and ventral), color pattern on all the coverts, Iris coloration, eye ring colorations, shank coloration, helmet coloration, neck ring coloration etc. Present study was also focused on differentiating the morphological variation between male and female guinea fowls N. meleagris. Altogether 30 specimens of N. meleagris were observed with morphological characteristics: humpedback appearance, slate grey plumage with numerous white spots throughout the feathers, neck short containing small feathers devoid of any ring, forehead and crown red in color, bill and casque horn colored. Iris was brown, whereas legs were dark grey. They were observed to possess short and long helmet, long reddish ear lobes and two wattles one from each side as well as pointed and short beak. Turkey birds were studied to possess feather-less head and contain long hair like out growths on the posterior side. The male guinea fowl was observed to possess large sized helmet or snood, whereas female possessed large sized helmet or snood. Male had large wattles and casque (horn on the top of head) that was relatively short or reduced in female. Body size of male was comparatively larger than female and alike variation was also recorded in size of neck and helmet.

# MORPHOLOGY AND DISTRIBUTION OF GREY PARTRIDGE (PERDIX PERDIX): FAMILY PHASIANIDAE INHABITING DISTRICT MIRPURKHAS, SINDH

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Present study was proposed to record the morphological characteristics and distribution of grey partridge (*Perdix perdix*) under family Phasianidae of order Galliformes in different study sites (talukas/subdivisions) including Mirwah, Digri, Tando Jan Muhammad, Jhuddo and Mirpur Khas city of district Mirpur Khas. Field surveys were conducted for observing the partridges from August 2018 to March 2019. The physical characteristics of grey partridges were examined with great focus on morphological variations among members of same species. Distribution of grey partridges was also emphasized during exploration of different study sites already mentioned above. Present study was also focused on arranging systematic of species in question of the study area "District Mirpur Khas" that was explored for the first time for the research on partridges. In this context, the existence of grey partridges was confirmed in Digri, Tando Jan Mohammad and Mirpur Khas city, however their presence was unconfirmed in vicinities of Mirwah and Jhuddo talukas. The results of present study showed the morphometric as followed: body weight (491.4 ±8.9), body length (28.7±6.1), head (3.1±5.0), beak (2.6±4.4), tail (4.4±3.8), torso (15.0±3.4), and toe (3.4±2.3). It was observed that the female *Perdix perdix* was smaller than male and also it was dull in color, whereas male was recorded as brightly colored and more attractive than female. It was recorded that

perdix perdix was abundantly distributed in most of the study area except Mirwah and Jhuddo talukas, where lack of agricultural lands (which are source of food and space) might have limited the distribution of grey partridge.

# EXTENT OF ILLEGAL TRADE ASSOCIATED WITH INDIAN PANGOLIN (MANIS CRASSICAUDATA) IN POTHWAR PLATEAU

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Pangolin is the world's most trafficked wild mammal, being illegally traded for its meat and scales. In the country, different poachers and hunters are involved in this illegal trafficking of the species to different parts of the country and abroad. The current study was conducted in the Pothwar Plateau, from September 2018 to July 2019 to collect data about extent of illegal trade of Indian pangolin and investigate its trade route. Data were collected from community using self-designed questionnaire along with animal market surveys. During the field/community survey, we also recorded the public perception about pangolin trapping technique, pangolin sighting areas, preferred season for hunting, preferred time for hunting, selling/buying points and Selling Price etc. In the second part of the current investigation, we dealt with under cover Market survey to unveil the Indian pangolin on sale in the markets of different cities of the country. This component of the study covered a total of 239 shops selling different wildlife species including mammals, birds and pet shops, local hakeem, street vendors and Pansari from five metropolitan cities, that is, Rawalpindi, Peshawar, Lahore, Karachi and Muzafarabad. Results showed that local hunters from study area were involved in illegal killing/poaching of Indian pangolin on the Pothwar Plateau. Out of 70 respondents from each district of the four districts of the Pothwar Plateau, maximum numbers of hunters were found in Chakwal District (n = 21 at 15 different locations), followed by Rawalpindi (n = 18 at 9 locations), Attock (n = 16 at 07 locations), Jhelum (n = 12 at 8 locations.. The maximum numbers of Pangolins trapped/poached were during the study period were reported from Chakwal District (n =160), followed by Attock (n =120), Jhelum (n = 35), and Rawalpindi (n = 35). Evidence collected showed involvement of nomads (gypsies) of the study area in capturing and selling of Indian pangolin on large scale. These nomads have been in direct contact with dealers from Lahore, Rawalpindi and Karachi cities Pangolin scales have been reported to be sold for Rs. 1000-2000/kg while live animal were sold for Rs. 50000-60000 through poachers and dealers. According to the record of the Punjab Wildlife and Parks Department, many people have been arrested who were involved in poaching / illegal trade of Pangolin from Jhelum, Chakwal, Rawalpindi and Attock during 2011-2017 and they were fined 30,000 PKR to 35000 PKR along with 6 months of imprisonment. During undercover market surveys, we identified few dealers in Thokar Niaz Baig, Lahore who are involved on large scale trafficking of Indian Pangolin meat and scales within the city and outside the city as well. These dealers sell the scales of Pangolin at different prices that may range from Rs.17000-18000/kg to the Chinese people who are residing in Defence Housing Authority (DHA) Lahore. But unfortunately, the Survey team was unable to find the scales and meat of Pangolin in herbalist shops, street vendors and local Hakeems. The large scale illegal trade of Indian pangolin for its scales from the Pothwar Plateau and its selling in different cities of the country and abroad demands immediate measures to be taken to stop this trafficking.

# CONSERVATION AND DIET PROVISION OF CHINKARA (GAZELLA BENNETTII) AND SPOTTED DEER (AXIS AXIS) IN THE CAPTIVE CONDITIONS OF LAL SUHANRA NATIONAL PARK

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Documented study was carried out to determine the conservation and diet provision of chinkara and spotted deer in the captive conditions of Lal Suhanra National Park. Chinkara and spotted deer are important ungulates in

Cholistan desert. Lack of suitable habitat and overhunting are the major factors of their decline in this region. Study was carried out during August 2018 to July 2019. 24 visits were made during the whole study period to monitor all the seasons. Under the captive conditions; habitat ecology, feeding behavior, population density and breeding behavior were estimated. Direct observation methods were used for observing all the activities of animals from dawn to dusk. Chinkara and spotted deer were better browser as well as grazers. They consumed flowers, pods, leaves, dry twigs and legumes of Acacia nilotica, Calatropis procera, Sacharum begalensis, Lycium barbarum, Syzygium cuminii and Moras alba present in the enclosure. They grazed on Sacharum begalensis whole year. Diet provided in the enclosure was green fodder like Jawar, Bajra and Jantar in the summer season. Berseem, Locern and Jawar were used for feeding as fodder in the winter season. Black gram provided whole year especially when green fodder deficiency was observed in the enclosure. About 800 gram per animal green fodder and black gram of about 20kg over all were used on daily basis. Total population mean value of Chinkara were observed 66.5±3.9. Total population mean value of Spotted dear were observed 15±1.04. In chinkara and spotted deer, mating was observed throughout the year. Mean value of chinkara fawn survival was 2.5±2.3. Mean value of spotted deer fawn survival was 0.41±0.51. For conservation of both species; area of enclosure should be increased, it will provide more wild conditions to the animals. Habitat should modify by planting indigenous species and shrubs in enclosure. Planting suitable browsing and grazing species in enclosure is helpful for habitat conservation. Enclosure fencing with metallic wires should be made to avoid the hunting and to avoid the escape of animals. Professional, trained and qualified staff should be employed in the enclosure.

# MORPHOLOGY, NESTING BEHAVIOR AND ECOLOGICAL IMPORTANCE OF *PASSER DOMESTICS* (HOUSE SPARROWS): FAMILY PASSERIDAE, SINDH, PAKISTAN

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Passer domesticus (House Sparrow) is a cosmopolitan bird species found almost everywhere except Antarctica. Its distribution is reported from different areas of Pakistan but its status is hidden in District Naushahro Feroze. Therefore, present study was proposed to record Morphology of P. domestics along with its nesting behavior and Ecological importance. In this perspective, extensive field surveys were conducted in different study sites including village Abdul Majeed, Village Muhammad Qasim, Village Sultan Mahesar, Village Ghanwar Mahesar, Village Roshan Mahesar, Village Qasim Sloangi, Village Hoat Magrio, Village Sher Muhammad Ganghro, Village Abdul Gafoor Mangrio, Village Mureed khan Lakho, and Villaage Sajan Mangrio of District N. Feroze. The morphological parameters including: Body weight, Bill coloration, plumage coloration, iris coloration, shank coloration, feathers and feet adaptations were analyzed for the identification and determination of morpho-taxonomic variation among members of same species. It was observed that male body weight varied from 24 to 39 grams (g), whereas female body weight was recorded within 21 to 37g. Male possessed dark spots on breast, whereas female breast exhibited white and grey spots. Males were dark and blackish grey, while females were light grey in colour. Bills of males were observed as dark in breeding seasons while females had whitish and grayish bills. Iris of male and female specimens was black, while body feathers in males were spotted with black colour and females had grey spots on their feathers. Color of the shank in males was usually black and sometimes whitish, while grayish coloration was observed in shanks of females. Feet were adapted for perching in both male and female house sparrow; however, male's feet had been adapted for digging as well. It was also observed that females were slightly smaller in size than male House Sparrows. The nests of house sparrow were observed to be made of straws, feathers and soft sticks of plant branches containing some leaves. P. domesticus was observed to exhibit harmful as well as beneficial role in ecosystem. It was observed as aggressive competitor with native birds, consuming large quantity of food, affecting biodiversity of other species of birds and an agricultural pest. P.domesticus was observed producing large quantities of feces and causing damage to the crops such as wheat, rice and sourgam severely. Their positive aspects include their role as biological pest control.

# ROAD KILLS, A SERIOUS ISSUE TO BE IN CONSIDERATION UNDER DEVELOPMENT OF ROAD-INFRASTRUCTURE IN FUTURE, A CASE STUDY IN DIR LOWER, KHYBER PAKHTUNKHWA, PAKISTAN

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Road executions are very common throughout the world. Vertebrates including mammals especially humans, birds, reptiles and invertebrates including insects, earthworms etc. are killed at an exponential rate throughout the world on highways and motorways. These kills are reported both by scientific community in scientific literature and by local, national and international print and electronic media. Research and survey before this one has not been led in the area to highlight the issue of road kills in the study area. As the government of Pakistan plans to include the northern areas of Pakistan in the list of tourist's hotspots, therefore we felt the need to highlight this issue for the government to be considered in the future plan, and for the people and scientific literature to blowout awareness throughout community in common. A total of 65 kills have been reported through this survey for a period of seven months in Lower Dir, Khyber Pakhtunkhwa. Overall, ten species were testified killed with highest percentage of domestic cat (*Felis catus*) 28 and lowest for goat (*Capra aegagrus hircus*) and fowl (*Gallus gallus*), for each of these percentage is 1% respectively. More animals were observed killed in hilly areas 67.69% than that of the plain areas 32.31%. The aim of this study is to highlight the issue of animal kills on roads due to traffic flow and to take a front picture of this hidden concern in Lower Dir, Khyber Pakhtunkhwa.

# DIVERSITY AND HABITAT ASSOCIATION OF SMALL MAMMALS IN QAZINAG FOREST RANGE, JHELUM VALLEY, AZAD JAMMU AND KASHMIR

# Aroosa Khalil<sup>1</sup>, Riaz Aziz Minhas<sup>1\*</sup>, Basharat Ahmad<sup>1</sup>, Muhammad Siddique Awan<sup>1</sup>, Khurram Shahzad<sup>1</sup>, Ghulam Ali<sup>1</sup> and Usman Ali<sup>2</sup>

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Small mammals comprised of an indispensable group of the vertebrates because of their diverse ecological roles and impact on the local economy of the area. However, perhaps due to their smaller size, they are often ignored in the scientific studies and conservation management strategies. Based on their indispensable importance, the current study was carried out to investigate the diversity and habitat associations of these mammals in Qazinag Forest Range, in Jhelum Valley, Azad Jammu and Kashmir. Twenty-three sites, belonging to five major localities and five major habitat types were surveyed for field data collection from July 2018 to July 2019. Direct and indirect methods including, visual encounter survey, trapping and opportunistic spotting methods were used to collect the data from different habitats. Besides, focal group discussion and literature review were used to supplement the data collected from the field. A total of 30 species of small mammals, belonging to 5 orders, 13 families and 27 genera were recorded from the study area. Most of the species was recorded for order Rodentia and Carnivora (9 species in each order) followed by Chiroptera (8 species) and Insectivora (03 species). Species richness and abundance was significantly different among different habitats ( $\chi^2 = 14.78$ , p = 0.005;  $\chi^2 = 655.97$ , p = 0.000), localities ( $\chi^2 = 11.37$ , p = 0.023;  $\chi^2 = 136.45$ , p = 0.000), elevations ( $\chi^2 = 13.82$ , p = 0.003;  $\chi^2 = 586.19$ , p = 0.000) and seasons (t = 1.69, p = 0.000). The highest species richness and diversity was recorded in agricultural fields (n=25; H'=0.365), followed by human settlements (n=18; H'=0.364) and forests (n=14; H'=0.234). The highest species richness and diversity was recorded in Nardajiyan (n=28; 388; H'=0.3610) and the lowest in Pandu (n=16; 157; H'=0.2584). Most of the species were found in lower and middle elevations (1200-2200 m) and a significant negative correlation between species richness (r = -0.718, p = 0.000) and species abundance (r = -0.633, p = 0.001) revealed a continuous decreasing trend of these attributes along the increasing elevations. There were significant variations in species richness and abundance in different seasons (t=1.69, p=0.000) and the diversity index was higher for summers (H'=2.4145) than the winters (H'=0.410). Although not severe, but several conservation issues such as unawareness in local community, habitat degradation, trapping, illegal trade and road killings were recorded in the study area. Findings of this study regarding the associations of different species to different habitats, localities and altitudes have uncovered important basic knowledge for conservation management planning of the area.

# ROOSTING ECOLOGY OF HOUSE CROW (CORVUS SPLENDENS) IN MUZAFFARABAD CITY, AZAD JAMMU AND KASHMIR, PAKISTAN

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Communal roosting refers to such kind of animal behavior where the grouping of various individuals of the same species takes place for specific interval of time. There are numerous assistances linked with communal roosting including reduced predation, decreased thermoregulatory demands, increased conspecific interactions and enlarged foraging ability. Understanding of roosting behavior can help the conservation management of the species under study. In present study we examined the characteristics of 23 different roosting sites and behavior of house crow in Muzaffarabad City of Azad Jammu and Kashmir. Patterns of house crow abundance (HCA) in various habitat features like elevation of the roosting sites (ELE), distance to nearest market or garbage dumping sites (DMG), distance to nearest open spaces (DOS), distance to nearest water bodies (DWB), distance to nearest buildings (DNB), distance to nearest major roads (DRD), number of substrates (NOS), Height of the substrate (HOS) and HOR (height of roost) were measured. A total of 10 roosting substrates including trees (08) and manmade structures (02) were used by house crow in the city. Among different parameters, two habitat features i.e. distance to the market and garbage (DMG) and distance to the nearby road (DRD) showed a significant effect on house crow abundance (HCA) in winter (DMG, p=0.006; DRD, p=0.057) and summer (DMG, p=0.009; DRD, p=0.037). In all the seasons, house crow started returning to their roosting sites from 45 minutes prior to sunset. It also reveals that birds return early during winter and late in summer. Vocalizations were found to decrease and finally stopped when all crows had returned to the roost. House crows were roosted close to one another and once settled, most of the crows tend to move towards the inner canopies. The study concluded that the house crow abundance in different sites were mainly determine by availability of nearby food sources in the form of carcass, house hold garbage, and rotten leaves.

### HUMAN THREATS TO CONSERVATION OF WILDLIFE DIVERSITY IN PAKISTAN

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Wildlife Diversity is the distinctive study related to various types of species existing in the eco-system. There should theoretically be a limit to the damage that humanity has inflicted on the planet — but clearly there is not .A very informative & thought provoking editorial, thread baring a serious global issue but having a profound impact to our fragile environment too. The ecosystem, of which humans are a destructive part, having existing for centuries undergoing changes due to instrusive human activities, have accelerated due to technolical advances of ever increasing world population. The rapid urbanization of the planet to accommodate demand of food & shelter, in the last 200 years has resulted in devastation of forests & green cover turning these into a concrete and asphalt jungles. Pakistan, one of the 5th most populous country, following the same trend has played havoc with its natural habitats. The threat of extinction to snow leopard up north, Ibex of Chiltan mountain, & Houbara Bustard in South, shows poor management of conservation of habitat. The reduction of forest area from 5.5% to 3.1%, during last 20 years, is

a cause of water shortages through climate change. Fast action needed to avert disaster. Current state of biodiversity conservation and management in countries all around the world had been discussed with an emphasis on Pakistan.

# MORPHOLOGY AND ECOLOGY OF HOUSE SPARROW (PASSER DOMESTICUS) OCCURS IN DIFFERENT AREAS OF PROVINCE SINDH, PAKISTAN

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The house sparrows are the imminent songbird. They are extended throughout the world today and belongs to the Class aves, Order passeriformes, Family passeridae and Genus *Passer*. They are indigenous to the Europe and larger part of the Asia. Accidentally they had been introduced in different domains of the world viz; Africa and Australia. In the course of present study, total of 63 species of the House sparrow were recorded in different localities of Sindh, Pakistan viz; Tando-Allahyar, Larkana, Mirpur Khas, and Jamshoro. The highest number of specimens were observed at Mirpur Khas whereas the lowest one at the remaining sites accordingly. Ecologically, observation reflects that they are associated with humans. They are observed in urban and rural areas but they avoid the grassland and desert areas. They mostly feed grains, seed and discarded food. All the recorded specimens were brought in the laboratory of vertebrate Biology Department of Zoology, University of Sindh, Jamshoro to identify the morphological parameters which includes; length and width of bill, head, wing chord, feet, plumage etc. Therefore, current study reflects the variation.

# SPATIO-TEMPORAL VARIATIONS OF AIRBORNE MICROFLORA IN AVIAN ENCLOSURES

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Since animal housing facilities are known to harbor a larger number of bio-aerosols, the air of five avian enclosures was sampled in two seasons with the help of a volumetric sampler. Air borne microflora was sampled for twenty minutes at each site depositing the air on Mixed Cellulose Ester (MCE) filters. The samples were then cultured for identification of potential bacterial species present in the air. A total of nine (9) species representing eight (8) genera were identified during the study with Gram positive bacteria being the most predominant form of airborne microflora including Micrococcus spp. Staphylococcus spp. and Streptococcus spp. to be the most abundant ones. Gram negative species included Enterobacter spp. and Pseudomonas spp. while E.coli and Salmonella spp. were also identified in a limited number of samples. The outcomes of paired sample t-test revealed a significant seasonal variation in colony forming units (p = 0.033) while eta squared statistics (0.7) indicated the effect size to be large.

### COMPARISON OF NESTING SITE PREFERENCES AND BREEDING ECOLOGY OF RED-VENTED BULBUL, BAYA WEAVER AND GREY BUSH CHAT IN SHEIKH BUDDIN NATIONAL PARK DERA ISMAIL KHAN, PAKISTAN.

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Nesting is a common phenomenon related to bird's reproductive behavior. Nests play a pivotal role in different developmental stages of bird's species including reproduction, protection, environmental measures, shelter and survival. Relation between a bird and tree/plant species is considered a major aspect of wildlife and biodiversity that makes nesting phenomenon a vital factor while studying the life of birds. The current study is conducted at Sheikh Buddin National Park. The total conducted study duration was from October 2018 to September 2019. Mostly birds breeding was observed in May to August. In the breeding season from December 2018 to august 2019. The dominant tree and shrub species include *Ziziphus jujuba*, *Ziziphus mauritiana*, *Acacia Nilotica*, *Cortaderia selloana*, and Sheesham *Olea oleaster*. Total thirty-nine (39) nests of 3 selected bird species Red-vented bulbul, Baya weaver and Grey bush chat were founded in which 18 active nest were r recorded at sampling sites in the study area. All the three birds nest were observed on *Acacia Nilotica*. The average highest nests height 5.06±0.43m were observed Red-vented bulbul. Maximum Nest n=8 of baya weaver bird observed. Grey Bush Chat successfully hatched 100%. The average incubation days for Red-vented bulbul 12.83±1.16, Baya weaver 12.71±0.75 and Grey bush chat 13.25±1.5 were recorded.

### THREATS OF MIGRATORY BIRDS IN PAKISTAN

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Migration is most important incident for animals to complete one annual life cycle. Many species of birds show local or long-distance migration in search of food or avoid harsh weather conditions as during winter season, large numbers of birds migrate from countries of central Asia and Europe towards Pakistan. Few birds migrate from Palearctic region afar from Himalayas are the sessional immigrants for breeding. Important migratory birds of Pakistan are flamingo, Falcons, Swans, geese, Waders, cranes and ducks. The large distance migration within breeding and wintering grounds results in evolution of certain traits in shore birds. Mostly bird's migration takes place from northern arctic region towards southern plains. Wintering season mainly spends in tropical areas where they inhabit 6-7 months and breeding occurs in temprte areas where they stay for two or three months. Siberian region birds migrate to oriental region through different fly zones in order to shun the perils of extreme cold annually in the winter season. In Pakistan Indus serves as a middle Asian fling route for the migratory birds that is joined to West Asian and East African fling routes. The disappearing trends of migratory birds are recorded from freshwater reservoirs of Pakistan and are mainly recognized to continued loss of wintering habitat, fragmentation, habitat modification, eutrophication, anthropogenic activities and illegal hunting. Agrochemical Contamination (Black-tailed godwit and Fish bird) Heavy Metal Pollution Contamination (Cattle egrets) Unsustainable Fisheries Practices (Ducks), invasive species (Little egret and Night heroine) and illegal hunting (Geese, Ducks, Siberian cranes and Bustards). Regular surveys for important migratory birds to assess to conserve important migratory birds, population and this is another step towards conservation of birds takes steps for the protection of their habitats and to prevent the illegal hunting, mitigating agricultural and industrial pollution, preventing water reservoirs from heavy metal poisoning are important considerations. Ceasing anthropogenic activities along with the mass awareness programs on electronic and print media could be effective conservation programs during the period of their stay in Pakistan.

# POACHING OF ASIATIC BLACK BEAR: EVIDENCE FROM SIRAN AND KAGHAN VALLEYS, PAKISTAN

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This paper assesses the vulnerability of Asiatic black bear to poaching in a key ecological zone in Northern Pakistan. Evidence about black bear hunting and cubs poaching were collected from Siran and Kaghan valleys of district Mansehra, KP Province through three different methods including participants' observations, interviews from key informants and structured questionnaire. We find local community members of both valleys to be active observers of hunting and poaching in the study area. The structured interviews with locals of the area revealed striking facts about ongoing trafficking by the hunters and traders of the neighboring districts (i.e. Kohistan and Battagram). The study identifies a tribe known as Maddi Khel in the neighboring district Battagram, the member of which regularly travel to the study area for the hunting of black bear, pheasants, gray goral and snow leopard. According to the interviews with the local people, hunters and traders in groups from 10 to 25 persons with their own poaching arrangements including guns, pistols and well-trained domesticated dogs are seen every winter. For hunters in the study area, the main target is the killing of female black bear with cubs because killing of mother allows them to poach their cubs. We also identify the routes-i.e Raam Gali, Door Gali, and Kunda Gali- through which hunters enter from the neighboring districts Kohistan and Battagram for the purpose of poaching. During each four months (December to March) season, on average, approximately 2 to 5 mothers are killed that carry 12 to 20 cubs. The small cubs have higher black-market demand in different cities in Pakistan that motivate the continuing of poaching in the study area. Despite the conservation claims by the government department in the province, the ground survey reveals severe risk to the population of black bear from hunting and illegal trafficking. The government's wildlife department staff is less equipped to tackle with hunters in winter due to the extreme weather conditions in the area and their inability to stay for longer time.

## TRADE & BUSINESS OF BIRDS IN LAHORE, PAKISTAN, THAT MUST BE FORBIDDEN BY LAW

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Birds are equally important Just like other animals in ecosystems, disturbance to this creature by human activities cause problems and its drawbacks on the environment including food web of the affected area can easily be seen in the issues about Vultures in Pakistan which are still the burning points for "Punjab Wildlife & Parks Department" and also causing a lot of expenses. The present summery was made to report a game that some unwise people are playing with exotic birds in Lahore for money. This includes a large number of birds belonging to same species kept in cages, they are selling them on roads without any concealment, and they set free a pair of bird when a customer pays for it. Conversation with many customers show that the purpose is some superstitious and religious blessings, further the amount per pair is so cheap that a common man can easily afford it. The most popular birds in this trade are *Hirundo rustica* (Barn swallow or Ababeel), *Motacilla alba* (white wagtail), many races of *Motacilla flava* (yellow wagtail) and

Motacilla citreola (citrine wagtail). According to IUCN Red List version 2019-3, the population trend of Hirundo rustica and Motacilla flava is decreasing, Motacilla alba is stable and that of Motacilla citreola is increasing. Moreover, all these birds are insectivore which can cause damage to the invertebrate fauna that may have importance in agriculture. Friendly commentary with these sales men revealed that there are well managed dealers who bring or purchase these birds on a large scale from out of the city and then deal them in small groups to these sales men so the overall profit is huge. It is a strong observation that in this business at different locations in Lahore a specific bird is most common at a particular time which represents large scale netting of that species from a rich habitat which may be a migration site. They use Hirundo rustica and Motacilla alba only in January and February while Motacilla flava and Motacilla citreola throughout the year. Two reports about the issue were written to the "Punjab Wildlife & Parks Department" and they replied that netting sites are not defined, further these birds are not protected in the wildlife Act of Pakistan so a serious action cannot be made. The concerned departments should take some sincere action and proper legislation is required to solve the problem.

## ASSESSMENT OF HEAVY METALS SUSCEPTIBILITY RISK TO MIGRATORY DUCKS NEAR RIVER RAVI

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Wetlands of Pakistan provide important wintering grounds to migratory birds. Due to different anthropogenic factors, these ecologically important habitats become contaminated by many pollutant including heavy metals that effect migratory birds as well. This current study has been planned to evaluate the effect of these toxins on winter migrants of Pakistan at River Ravi. To accomplish this task four heavy metals Cd, Cu, Cr, and Pb were detected from vital organs of migratory ducks (Gadwall, Pochard, Shoveller, Teal and Mallard) by Atomic absorption spectrophotometry. The histopathological examination of these organs has also been done to access damage caused due to selected heavy metals. The ducks organs were obtained from hunters at River Ravi belt. Results showed that the major organs with most significant changes due to heavy metal toxicity were kidney and liver respectively. Among the metals, highest concentration was detected for cadmium in liver of Shoveller that was 61.5µg/g whereas concentration of chromium was below detection limit (BDL) in all tested samples. The highest concentration of lead was detected in liver of shoveller i.e. 5.73μg/g and gadwall showed lowest value of lead that was 1.14μg/g. The pattern of concentrations of lead (Pb) was Shoveller > Mallard > Teal > Gadwall > Pouchard. As far as the cadmium (Cd) is concerned pattern of concentration was Shoveller > Gadwall > Mallard > Pouchard > Teal. The highest concentration of copper (Cu) was obtained from pouchard liver 20.7µg/g and lowest was in liver of shoveller i.e.12.7µg/g. The concentration pattern of copper was Gadwall > Pouchard > Shoveller > Teal > Mallard. The most affected organ was kidney. Histopathological sections of the kidney showed the presence of renal tubular cells in the lumen of tubules, severe focal area of coagulate necrosis and tubular degeneration was also evident. The second most effected organ was liver. Where severs hepatocellular and hydropic degeneration with extensive photolytic regions within cytoplasm was noticeable.

# STATUS AND HABITAT USE OF GREY FRANCOLIN (FRANCOLINUS PONDICERIANUS) IN PIR LASURA NATIONAL PARK, AZAD JAMMU AND KASHMIR

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A study on the status and habitat use of Grey Francolin (*Francolinus pondicerianus*) was conducted in Pir Lasura National Park (PLNP) Azad Jammu and Kashmir (AJ&K). Pir Lasura National Park, situated at 33.639204 °N and

73.846664 °E, with an area of 56.25 km2, falls in distribution range of grey francolin species. The area was surveyed from February 2016 to December 2016 between altitudinal ranges of 1200-1800 m above sea level (asl). Study area was divided into five main localities with nine sub-localities and systematic surveys were conducted in each sub-locality. For distribution, thorough surveys were conducted to collect direct and indirect evidences of its presence. Population was estimated using line transect method and selected transects were surveyed early in the morning and before evening. During the survey, geographical coordinate, elevation, weather conditions and general habitat conditions were recorded. The result indicates that the grey francolin was present in all selected localities of the study area. Overall population density of grey francolin in the study area was 3.82 birds / km2. The highest density was recorded in sub-locality Sairi khandhar as 5.5 birds / km2 and lowest in Lanjot 2.06 birds / km2. Habitat us was determine through vegetation surveys using quadrate method of different sizes for trees, shrubs and herbs, along each transect as per suitability of the habitat. Pinus roxburgii, Dalbergia sissoo, Acacia nilotica, Punica granatum, Ziziphus jujuba, Cynodon dactylon and Heteropogon contortus were dominant species of grey francolin habitat. A total of 58 plant species were recorded in the habitat including trees (n=18), shrubs (n=14), and herbs and grasses (n=26). The overall flora of the habitat was dominated by third strata of vegetation consisting of herbs and grasses (44.82%) followed by trees (31.03 %) and shrubs (23.13%). Data of the study will be help maintain population, improve habitat and conserve grey francolin in Pir Lasura National Park as well as in Azad Jammu and Kashmir.

# POPULATION DENSITY AND HABITAT ASSOCIATION OF BLACK FRANCOLIN (FRANCOLINUS) IN MARGALLA HILLS NATIONAL PARK, ISLAMABAD

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The study on population density and habitat association of Black Francolin (Francolinus francolinus) was conducted in Margalla Hills National Park, Islamabad from March to September 2018. The objective of this study was to estimate the population density and analyze the habitat association of Black Francolin in the study area. The study area was divided into four study sites viz., Site-I (Nurpur Range), Site-II (Saidpur Range), Site-III (Golra Range) and Site-IV (Shah-dara Range)..Black Francolin's population density data was collected through direct sightings using line transect method. In each study site four transects were laid down with a length of 500 m and width ranging between 20 to 40 m. The population data was gathered at each study sites every month for 3 consecutive days at both morning and evening. Population density of Black Francolin in the study area was estimated as 0.8 birds/ha. The maximum population density was noted in Study site-II (1.2/ha), followed by study site-I (1.07/ha) and study site-IV (0.52/ha), and low population density was observed in Study site-III (0.41 birds/ha). For analysis of habitat use, vegetation survey of study area was conducted in four study sites using quadrate method. Ten quadrates were taken in each study sites. Overall 40 transects were taken in the whole study area. Out of 30 plant species noted from the habitat of Black Francolin, 8 trees, 11 shrubs, 8 herbs and 3 grasses were noted. The dominant tree species at the four study sites (I, II, III and IV) were Acacia modesta, Pinus roxburghii, Bauhinia variegata, and Cassia fistula. The major shrub species were Lantana camara, Dodonaea viscose, Carissa opaca, and Buxus papillosa and main herb species were Datura innoxia, Lathyrus aphaca and Oxalis corniculata. The grass species were Heteropogon contortus, Cynodon dactylon, and Apluda mutica.

# HABITAT CHARACTERISTICS AND POPULTION DENSITY OF MUSK DEER (MOSCHUS CHRYSOGASTER) IN ASTORE VALLEY, GILGIT-BALTISTAN

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Musk deer (*Moschus chrysogaster*) belongs to the family Moschidae and genus Mosshus. Musk deer are small solitary ruminants that inhabit the forested and alpine scrub habitats of mountains in Asia. Out of seven species of Musk deer only Himalayan Musk deer is found in Pakistan. It is now distributed in patches of its previous continuous

range in the Himalayas. In Pakistan it is critically endangered. Musk deer population has experienced strong depletion, attributed to the loss of habitat, poaching and fragmentation. The present study was conducted to determine habitat characteristics and population density of Musk deer in Astore valley Gilgit-Baltistan. Line transect method was used for estimating its population. The results showed that the Population density of Musk deer was 1.01 animals/km² occurring between 2938 m to 3492 m elevation. Quadrate method was used for habitat analysis of Musk deer in the study area. It was noted that conifer forest was the preferred habitat of Musk deer. Majority of the animals were distributed between 2900 m to 3100 m elevation. From the presence of fecal pellet groups it was recorded that 82% of fecal pellet groups were present in forest land, 16% were in shrub land and only 2% fecal pellet groups were present in herbs or grass land. A total of 28 plants were identified in its habitat, of which 6 were trees, 6 shrubs and 16 were herbs. Dominant trees of the study area included *Betula utilis* (IVI=69.44), *Picea simithiana* (IVI=63.62) and *Abies pindrow* (IVI=59.36). Dominant shrubs included *Rosa maschata* (IVI=60.39) and *Hieracicim herisoides* (50.83). Among herb layer *Viola spp* (IVI=22.19) and *Cirsium species* (IVI=22.19) were dominant in the Musk deer habitat. Results of the study will assist in conservation of the species and its habitat in the study area.

# POPULATION DEMOGRAPHY OF SELECTED HERBIVORES IN THE PRESENCE OF PREDATORS IN ARABIAN WILDLIFE PARK OF SIR BANI YAS ISLAND, UNITED ARAB EMIRATES

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Predators are known to directly impact the population size and dynamics of the prey species. These effects include reduction in population size through predation as well as by changing the demography of the prey. Sir Bani Yas Island is a wildlife reserve having more than 16000 individuals mainly from herbivore species. These individuals rely on supplementary feed along with browsing, as their population exceeds the carrying capacity of the island. In the current study we recorded the population density and dynamics of the selected prey populations (Blackbuck, Sand gazelle, Mountain gazelle, and Axis deer) between 2017 and 2019. The data was collected annually through drive count method. The average population for three years was 346.67±36.02, 533±86, 1182±157, and 5222±202 individuals per year for Axis deer, Blackbuck, Mountain gazelle, and Sand gazelle, respectively. The average male population of Axis deer, Black Buck, Mountain Gazelle and Sand Gazelle was 336±37, 165±45, 538±59 and 1532±154 respectively. The average female population of Axis deer, Blackbuck, Mountain gazelle and Sand gazelle was 10±2.52, 327±90.39, 574±87 and 3212±157 respectively. The average juvenile population of Blackbuck, Mountain gazelle and Sand gazelle was 98.3±55.08, 70±23.58 and 492±125 respectively.

# PREY PREFERENCE AND HUNTING BEHAVIOR OF ARABIAN STRIPED HYAENA (HYAENA HYAENA SULTANA) AT SIR BANI YAS ISLAND, UNITED ARAB EMIRATES

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The Arabian striped hyaena (Hyaena hyaena sultana) are nocturnal carnivores of Hyaenidae family with five subspecies based on geographical variations and are listed as near threatened (NT) under IUCN Red List of Threatened Species. Sir Bani Yas Island in UAE is home to 13 individuals of Arabian striped hyaena. Sir Bani Yas Island has 41 square kilometres protected Wildlife Park, inhabited by approximately 16000 animals belonging to 32 species of various herbivores, carnivores, reptiles, and birds. Striped hyaena is reported mainly as scavenger but occasionally hunt prey up to the size of a barbary sheep (Ammotragus lervia). In the current study, we aimed to record the prey preference of Arabian striped hyaena when abundant prey population and variety is available in space limited wildlife reserve. The data on prey species, sex, age, degree of consumption, date and time of kill, and kill

location was collected between 2014 and 2016, through direct and indirect observations. The most abundant prey species was Arabian sand gazelle (*Gazelle subgutturosa marica*) comprising about 79 % of all predated species, followed by 6 % each for both blackbuck (*Antilope cervicapra*) and mountain gazelle (*Gazella gazella cora*), axis deer (*Axis axis*) 5 %, and 2 % each for barbary sheep and Arabian tahr (*Arabitragus jayakari*). Arabian striped hyaena predated more males (55 %) compared to females (45%). Most of the kills were recorded during the early morning (56 %) followed by 21 % at night, 13 % during the afternoon, and 10 % in the evening. Moreover, adult prey was preferred by Arabian striped hyaena (89 %) as compared to sub-adult (6 %) and juveniles (5 %). The preliminary results of the current study suggest that Arabian striped hyaena prefer to hunt if there are abundance and diversity of prey species around them.

## 5. BIODIVERSITY

# ASSOCIATION OF ALT AND AST WITH DYSLIPIDEMIA IN NEWLY AND KNOWN TYPE 2 DIABETES

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Liver plays a paramount role in regulation of glucose homeostasis; many studies have been reported that the liver diseases play a pivotal role in both morbidity and mortality of T2D patients. To our knowledge, limited studies have demonstrated the relationship of ALT with diabetes, while no such study in this part of the world exist for AST in relation to T2D. The aim of this study is to evaluate the association of ALT and AST with dyslipidemia in newly and known type 2 diabetes (T2D). This prospective clinical study was planned at university of Karachi with collaboration of Baqai Institute of Diabetology and Endocrinology. Subjects were categorized into four groups; Group II: 25 healthy with normal glucose tolerance, Group II: 25 newly diagnosed diabetes (NDD), Group III: 35 known T2D with <5 years duration and Group IV: 65 known T2D with >5 years duration. Data was collected on predesigned questionnaire. Blood samples were analyzed using laboratory techniques. Total 150 subjects had mean age 52.94±10.64 years. In subjects with normal glucose tolerance, only AST was significantly negatively correlated with FBS (r=-0.406, p=0.054). In NDD, ALT was positively significantly correlated with triglycerides (r=0.418, p=0.04), while both AST and ALT were significantly negatively correlated with HDL. In <5 years duration of T2D, AST was negatively correlated with HbA1c. In subjects with >5 years duration of T2D, ALT was significantly negatively correlated with LDL (r=-0.292, p=0.025) and HDL (r=-0.291, p=0.28), on the other hand, AST was significantly negatively correlated with total cholesterol (r=-0.323, p=0.014), LDL (r=-0.453, p<0.0001) and HDL (r=-0.318, p=0.016). Elevated ALT and AST with deranged dyslipidemia were found in known T2D as well as in NDD subjects. Routine screening of ALT and AST with lipid profile in T2D and NDD subjects may assists early detection of liver abnormalities and to arrest the progress of disease.

# DIVERSITY AND SYSTEMIC OF AMPHIBIAN FAUNA OF FAMILY RANIDAE AND BUFONIDAE, DISTRICT NAUSHAHRO FEROZE, SINDH, PAKISTAN

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Amphibians are tetrapod vertebrates that inhabit a wide variety of habitats including terrestrial, fossorial, arboreal or freshwater ecosystem. Class Amphibia is divided into three subclasses: Lepospondyli, Temnospondyli and Lissamphibia; two former subclasses are extinct, while subclass Lissamphibia represents three orders i.e. Anura, Urodela and Apoda. In Pakistan, there is no evidence of existence of Caudata/Urodela and Gymnophiona/ Apoda, however Anura is single existing order of class Amphibia. Order Anura is declining fast due to variety of reasons including contamination, climate change and over-exploitation. In this context, present study was conducted to record the diversity and systematic of frogs and toads under order Anura in district Naushahro Feroze. Thorough exploration carried out from August 2018 to October, 2019 recorded the distribution of diverse anurans (tailless amphibians). The morphometric of different body parts: body weight, body length, limbs length, eye diameter and tympanum diameter were measured along with observation of other morphological parameters including Dorsal and ventral body color, presence/absence of mid-dorsal line, head wide/narrow, snout wide/pointed, loreal shape, canthus shape, fingers

pointed/not, inter-orbital space narrow/wide, tibio-tarsal articulation shape, toes blunt/pointed using equipment of scientific grade. The systematic of each species was settled using identification key and relevant literature. The observed amphibian specimens were identified as two species "Hoplobatrachus tigerinus and Euphlyctis cyanophlyctis" of true frogs (Family Ranidae) and one species i.e. Bufo stomaticus of true toads (Family Bufonidae). It was confirmed that there was no existence of any other species of frogs and toads in the study area and also there was no morpho-taxonomic variation within members of distinct species. Though minor variations in body coloration and pattern of patches were observed that may occur due to different quality of amphibian habitats in different water bodies such as ditches and ponds. Existence of three anuran species falling in three genera (Genus Hoplobatrachus, Euphlyctis: Family Ranidae and Genus Bufo: Family Bufonidae) was recorded abundantly distributed throughout the study area during whole study period.

# DISTRIBUTION, MORPHOLOGY, SYSTEMATIC AND DIVERSITY OF FAMILY LEPORIDAE (RABBITS AND HARES), ORDER LAGOMORPHA, DISTRICT NAUSHAHRO FEROZE, SINDH, PAKISTAN

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Leporidae is one of the two families of order lagomorpha which includes rabbits and hares. It consists of about 62 species with eight different genera classified as rabbits including Brachylagus, Bunolagus, Nesolagus, Oryctolagus, Pentalagus, Poelagus, Romerolagus, and Sylvilagus, while genus Lepus represents Indian hare (Lepus nigricollis), Woolly hare (Lepus oiostolus) and Cape hare (Lepus capensis). Present study was proposed for recording distribution and diversity of Leporids with great focus on settling their morphology and systematic classification in an area "District Naushahro Feroze" where no relevant study was previously conducted. In this context, field surveys were conducted at different study sites such as village Yousif Rajper, village Malhar Khan Rajper, village Cheeho, village Rajab Khan Rajper, Padidan City, village Nibhao Khan Rajper, village Niza mu Din Arain, village Chuttal Khan Rajper, village Maman Khan, village Hassan Khokhar, village Khundha Khai, village Yaquib Tunia, village Baksho Khan Rind. Specimens of rabbits and hares were examined for morphometric i-e body weight, body length, length of fore limbs and hind limbs, tail, ears, coloration of fur and Iris using the material and method of scientific grade. The systematic of each species was determined using identification key and relevant literature. The existence of two species viz: Lepus capensis (cape hare/brown hare) and Oryctolagus cunicullus (domestic rabbit) was recorded randomly distributed in the study area. The morphometric of Lepus capensis was measured as followed: body weight: 1733.3±602.2, body: 19.8±2.2, tail: 3.5±0.8, eye: 1.2±0.9, hind limbs: 2.2±0.1, fore limbs: 1.3±0.1 and ears: 4.5±0.5. The body parameters of Oryctolagus cunicullus were recorded as: body weight: 1261.7±21.4, body length: 16.7±1.0, tail: 3.2±1.2, eye: 1.1±0.1, hind limb: 2.2±0.2 fore limb: 1.4±0.1, ears: 4.0±0.3.

### SURVEY ON POLLUTION EFFECT ON NESTING BEHAVIOR OF GREEN SEA TURTLE AT HAWKE'S BAY, KARACHI

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Being one of the biggest species of sea turtle in the ocean, *Chelonia mydes* is the most unique yet endangered species of the order Testudines cited by IUCN, it lives in the big wide ocean but lays eggs on beaches of all around the world. The turtle beach of Hawks bay Karachi is one of the nesting areas of green sea turtles. Green sea turtles are about 2 to 6 feet in length and they weighs about 200kg. At Hawke's bay Karachi, pollution, physical barriers, and predators are the noted cause of defaunation of the *C.mydes*. During the time period of 4 months (August –

November). Many major surveys were conducted within the radius of 3km during late afternoon to late night at the beach of hawks bay Karachi. The surveys provide the updated information on pollutions' effects on nesting behavior of green sea turtles, by the help of Marine Turtle Conservation Unit, the morphology of the Green Sea Turtle and there nesting behavior was thoroughly studied. About 27 nests were observed in which 11 were totally destroyed by humans and predators. Total number of turtle observed was 28 in which 8 went without nesting due to the too bright lights and huts present at the beach. The total number of nesting recorded was 24 clusters in which 14 were filled with plastic bags. The morphology of each turtle was attentively observed, about 10 turtles that came to the beach were between the sizes of 2-4 feet in length and about 1 feet in width, 15 of them were about the size of 4 to 6 feet in length and 2-4 feet in width they were mostly recorded while performing nesting ritual During the surveys, it was observed that the pollution was filling up the nesting areas with plastic bags which was inhibiting the female C.mydes from performing nesting in that area. Along with coastline the hawks bay beach is full of huts which are built for tourists, they are consuming the major areas of the nesting site of C.mydes, acting as a barrier between turtles and their nesting site, this is majorly effecting the rate of nesting and incubation of the eggs, as they are forced to nest near the tides, the probability of their hatching is very low due to temperature fluctuation. Many turtle were found dead due to chocking on plastic materials (mainly water bottle caps and plastic bags) at the coastline of Hawke's bay beach. The aggressive defaunation of specie Chelonia mydes is causing imbalance in the oceans all around the world, as green sea turtle plays an indirect yet important role to sustain the biodiversity of the ocean. With all this aggressively increasing pollution and built barriers, Chelonia mydas is been haunted by human greed's terror. Many steps have been taken by the officials to conserve this striking species from the aggression of humans' greed but the defaunation of *Chelonia mydas* is much greater than their conservation.

# SPECIES COMPOSITION AND DIVERSITY OF MACRO-BENTHIC POLYCHAETES IN MANGROVE SWAMPS NEAR PORT QASIM, KARACHI

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The polychaete assemblages at Russian beach mangrove swamps, near Port Muhammad Bin Qasim, Karachi were studied from January to June 2018 to analyze community structure and species diversity. Sediment samples were collected from the beach by a quadrat (30x30x10 cm). Nine samples were collected in each month for six months i.e. 54 samples altogether. Sediment samples were sieved through 1 mm mesh and the polychaetes were sorted out under stereomicroscope. A total of 20 polychaete species, belonging to 16 families were recorded. Families Ampharetidae, Capatellidae, Cirratulidae and Nereididae were represented by two species each. Other 12 families were represented by one species each. *Melinna aberrans* Fauvel, 1932 was the most abundant species (38%), followed by *Heteromastus filiformis* (Calaperede, 1864) (22%) and *Neanthes glandicincta* (Southern, 1921) (20%). These three species accounted for 80% of the total polychaete worms collected. The highest number of species were found in May and June (11 species in each month), whereas the lowest number of species were recorded in February and April (9 species in each month). Stress-Predictability modeling, based on values of Shannon diversity (H') and Pielou's evenness (J) was applied to established the level of stress existing in the study area with regard to polychaete species.

## A REVIEW ON AGRICULTURAL LANDSCAPES: FIELD MARGIN HABITATS AND THEIR ROLE IN SUSTAINING BIODIVERSITY

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Agriculture dominates the world terrestrial area, varying with cropping system, topography and intensity of management. Agricultural landscapes consist of mosaics of farmland and remnant natural habitats of woodlots,

shelterbelts and riparian zones that may offer an opportunity to conserve biodiversity while maintaining food production. In this paper, we review on crop field margins habitat which are important for enhancing biodiversity and associated ecosystem services on farms for long term agricultural sustainability. Agricultural fields are surrounded by fallow land, which provide suitable sites for shelter and breeding for a variety of wildlife including avian, reptilian and small mammalian species while shrubs, grasses, annual weeds and forbs may provide supplementary food. Field margins are also particularly valuable for conservation of plant diversity, livestock grazing, provide fuel wood, enhanced crop pollination, arthropod pest control, providing shelter for crops, particularly as windbreaks, reducing soil erosion by wind and water quality protection in agricultural landscapes. Agricultural operations, such as fertilizer and pesticide application, can have adverse effects on the margins. The biodiversity of the margins may be of particular importance for the maintenance of species at higher trophic levels, notably farmland birds, at the landscape scale. Field edges can also contribute to the sustainability of production, by enhancing beneficial species within crops and reducing pesticide use, though further research on the predictability of these effects is needed. Despite the benefits of field margin habitats and financial support to farmers, few landholders (growers/farmers) adopt biodiverse field edges. One reason is the perceived risk of increased damage from wildlife (especially rodents) using these habitat plantings and the potential for transfer of zoonotic enteric foodborne pathogens to human food crops by fecal contamination which pose food safety risks. The review revealed that agricultural landscape homogenization has detrimental effects on biodiversity and key ecosystem services. The review suggested that increasing agricultural landscape heterogeneity by increasing semi-natural cover can help to mitigate biodiversity loss. However, the amount of semi-natural cover is generally low and difficult to increase in many intensively managed agricultural landscapes. The field margin strips offer a practical means of providing on-farm biodiversity and enhancing more environmental and sustainable production.

# EXPLORING THE HERPETO FAUNA OF TOTALAI GAME RESERVE DISTRICT BUNER, KP

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During the current study 13 species of Reptiles and 4 species of Amphibians were reported. The total 17 species of Herpeto fauna were reported from Totalai Game reserve KP, Pakistan. The reported species belonging to 15 genera. The amphibian species *Duttaphrynus melanostictus* and *Duttaphrynus stomaticus* are most abundant and cosmopolitan species of herpetiles and mostly found at cultivated lands, human settlements and natural habitats of Totalai Game reserve. *Euphlyctis cyanophlyctis* is the common frog of current research area while *Sphaerotheca breviceps* seemed as the rarest species. *Hemidacctylus flaviviridis, Crytopodian kachhense, Calotes versicolor, Eutropis dissimilis* and *Varanus bengalensis* are the most common and abundant species of Totalai Game reserve. *Hemidacctylus flaviviridis, Crytopodian kachhense* are highly restricted to human settlements while *Varanus flavescens, Naja oxiana, Platyceps vantromaculatus* and *Varanus bengalensis* are found in natural and as well as urban habitats. *Calotes versicolor, Laudakia himalyana, Scincella himalayana, Eutropis dissimilis, Eublepharis macularis, Bungarus caeruleus* and *Ptyas mucosus* are limited to natural areas. The specie richness calculated having value 7.28. The Shannon Winner diversity index (H) have high value 2.192 and Simpson index (1-D) is 0.86. the herpeto diversity at Totalai Game reserve are some what evenly distributed having Evenness value (E) 0.53 and Equitability (J) 0.77. Fisher's alpha was also used because it includes rare species, giving them the same value regardless of their abundance and has higher biological significance having value 2.71.

# DIVERSITY, ABUNDANCE AND NICHE OVERLAP AMONG EARTHWORMS OF MAIZE (ZEA MAYS), RICE (ORYZA SATIVA) AND MILLET (PENNISETUM GLAUCUM) CROPS OF DISTRICT JHELUM

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Earthworms are important component of agroecosystem soil. They are also called as engineers of ecosystem. By the breakdown of organic matter earthworms liberate nutrients for utilization by bacteria and fungi. The current study was conducted to determine the diversity, abundance and niche overlap among earthworms in maize, rice and millet crops of district Jhelum. The samples were collected from July 2016 to October 2016 fortnightly. Sampling was done by selecting quadrat method along with the use of digging and hand picking method. A total of 360 specimens were taken from maize, rice and millet crops from Dharyala Jalip locality of district Jhelum. Keys provided by Stephenson (1923) and Bhatti (1962) were used to identify the specimens preserved in ten percent solution of formalin. For the analysis of data Shannon Weiner diversity index was applied, T-Test was used for crop wise comparison of fauna. Correlation was applied to find relationship between fauna and environmental factors and SPOVRLAP used for niche overlap using Bas Software. Maize crop had high diversity index H'=2.133 as compared to rice and millet, in rice diversity index was H'=1.931 and in millet diversity index was H'=1.872. Correlation coefficient for temperature and moisture showed significant results while pH showed non-significant results as for as diversity of earthworms was concerned. T- Test showed non-significant results for comparison of diversity in all the three crops i.e. maize, rice and millet. In specific niche overlap P. suctoria and P. houlleti were dominant species with niche overlap 0.990 in maize, rice and millet crops. In maize, rice and millet crops general niche overlap was GO = 0.819.

# ANTHROPOGENIC PRESSURE IN TERMS OF FOREST UTILIZATION AS FUEL WOOD AND TIMBER BY COMMUNITY OF MDNP NEELUM, AZAD JAMMU AND KASHMIR, PAKISTAN

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An increase in human population and anthropogenic pressure imposes serious threats to biodiversity and even many species are at verge of extinction. A global goal was set to establish the protected areas (PAs) for conservation of natural resources. Pakistan is also placed among those countries where human population growing rate is high and there is severe threat to biodiversity, however, there are number of National parks are established, where conservational plans are executed. Musk Deer National Park (MDNP) Gurez the protected area (PA) located in District Neelum, Gurez valley Azad Jammu and Kashmir, Pakistan. Total of 29 surveys, and questionnaire (n=576) were carried out to study the anthropogenic pressure in terms of utilization of forest as fuel and timber in MDNP. Data suggest that total of n= 22784 human population settled in 2618 household during the whole year in different parts of the MDNP. Moreover, a vast number of people (n=442 families) along with their livestock from outside the park move to hilly area during summer (May-August) in different region of MDNP. These people rely on forest for burning fuel. As fuel utilization vary in different month of year so, months are categorized into three classes May-August followed by September-November and December-April. It has been estimated that single family (average of 5 members) utilize an average total of 2043 kg/capita/year. The value of wood consumption was estimated an

average of 5544.822 metric tons in whole year for 3060 families inhabiting in MDNP. The average fuel wood consumption for 3060 families were recorded as 1358640 kg/capita/year (1358.64 metric ton) from May-August, followed by 3102330 kg/capita/year (3102.33 metric ton/year) for the month of December-April and 1083.852 metric tons fuel utilized by whole families (2618) during the month of September-November. Maximum average fuel wood consumption 9.3kg/capita/day was estimated among the families which living near forest less than 1km followed by 5.1 kg/capita/day those living less than 5 Km from forest. However, those families which living at distance 5 to 10 Km from forest consume minimum quantity 3.9 kg/capita/day wood as fuel. Forest uses as timber was also estimated by counting and catalogue the buildings, out of them 114 houses, guest house (n=22) and animals shed (n=128) were constructed during the year of 2013-2018. Maximum wood (42.02m³) is consumed for guest house construction followed by local house (22.08m³) while minimum wood (4.1m³) for construction of animal shed. Large quantity of wood is consumed for house construction (2517.12 m³) followed by guest houses (924.44 m³) and 524.8 m³ was used in animal shed.

### PROSPECTS OF AMPHIBIAN RESEARCH AND CONSERVATION IN PAKISTAN

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Despite concerns about global decline in the populations, amphibians do not find any place in wildlife conservation and management decisions, policy making and research agendas in Pakistan. We are conducting detailed studies and conservation awareness program on amphibians of Pakistan for the first time. During the first phase of our projects, we recorded eight anuran species from sub-tropical scrub and pine forest (District Rawalpindi and Islamabad Capital Territory). Our results showed that pine forest featuring high altitude and natural freshwater streams with low water temperature (<13 °C) had a relatively higher anuran endemism. Space use data suggested high site fidelity by endemic anurans (Hazara Torrent Frog, Murree Hills Frog). Increase in the atmospheric/ water temperature or water withdrawal from the streams by local community could seriously impact populations of these species. The species might be forced to perform over land migration through the forest to occupy nearest streams which are situated at a distance difficult to travel by amphibians or upstream migration that would require considerable energy reserves and may cause stress in the individuals. The data on impacts of climate change on amphibian species inhabiting northern parts of the country are lacking. However, it is feared that these species might face similar consequences as that of amphibians found elsewhere in the world. We believe a few anuran genera require detailed taxonomic studies, and that molecular taxonomy would add more species to known anuran diversity. We have suggested options for education, research, conservation, management and stewardship of amphibians of Pakistan.

# ABSTRACTS PRESENTED AT THE FIRST VIRTUAL CONGRESS OF ZOOLOGY

(40<sup>th</sup> Pakistan Congress of Zoology – International) December 17-19, 2021

## **SECTION - V**

## FISHERIES, ECOLOGY, WILDLIFE, FRESHWATER BIOLOGY AND MARINE BIOLOGY

### 1. FRESHWATER BIOLOGY AND FISHERIES

# STUDY OF SHARE OF VISCERAL MASS IN TOTAL BIOMASS, LENGTH-WEIGHT RELATIONSHIP AND TOTAL LIPIDS IN ROHU (*LABEO ROHITA*) CULTURED UNDER HIGH STOCKING DENSITY CONDITIONS OF IN-POND RACEWAY SYSTEM

#### Wajiha Annum' Shafaq Fatima and Hira Arshad

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In-pond Raceway (IPRS) is a recent technology introduced for first the time in Pakistan (2019) in order to increase aquaculture production. IPRS is the combination of raceway technology, cage culture, recirculating aquaculture and pond culture. Introduction to IPRS in countries like Pakistan where water shortage is a main issue would be a revolutionary step. High stocking density in IPRS may cause stress in fish and affect growth, immunity and survival. Present study aims at investigating the effect of high stocking density on body length, body weight, length-weight relationship (LWR), condition factor, hepatosomatic index (HSI), viscerosomatic index (VSI) and lipid content. It will provide a baseline data for future studies on IPRS in Pakistan. In present study, there were total 8,000 rohu (initial wt. 250.00 ± 7.5). The trail period is from August till November, 2020. Samples of five fish were collected after the time interval of 30 days and 50 at the time of harvesting (30 November). After sampling the growth parameters (Total body length, Total body weight, Condition factor and HSI) length-weight relationship, viscerosomatic index (VSI) and lipid content were analyzed. Total body length (cm) of Rohu was observed within the range of  $35.28 \pm 0.34 - 38.79 \pm 0.94$  cm, Total body weight (g) was observed within the range of  $826.00 \pm 77.99 987.00 \pm 63.31$  g, Condition factor was observed within the range of  $1.51 \pm 0.09 - 2.06 \pm 0.26$  %, Hepatosomatic index was observed within the range of  $1.01 \pm 0.00 - 1.28 \pm 0.07$  %. Viscerosomatic index was observed as  $2.00 \pm$  $0.13 - 6.75 \pm 0.13$  %. Regression coefficient (R<sup>2</sup>) value of Length-weight relationship was 0.466 while value of coefficient b was 1.139. In conclusion, high stocking density in IPRS did not affect the growth of Rohu.

# SHARE OF VISCERAL MASS IN TOTAL BIOMASS, LENGTH-WEIGHT RELATIONSHIP AND TOTAL LIPIDS IN GRASS CARP (*CTENOPHARYNDODON IDELLA*), CULTURED UNDER HIGH STOCKING DENSITY CONDITIONS OF IN-POND RACEWAY SYSTEM

#### Hira Arshad, Shafaq Fatima, Wajiha Annum

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In-pond Raceways (IPRS) is a recent technology introduced for first time in Pakistan (2019) in order to increase aquaculture production. IPRS is the combination of raceway technology, cage culture, recirculating aquaculture and pond culture. Introduction to IPRS in countries like Pakistan where water shortage is a main issue would be a

revolutionary step. High stocking density in IPRS may cause stress in fish and affect growth, immunity and survival. Present study aims at investigating the effect of high stocking density on body length, body weight, length-weight relationship (LWR), condition factor, Hepatosomatic index (HSI), Viscerosomatic Index (VSI), and lipid content. It will provide a baseline data for future studies on IPRS in Pakistan. In present study, there were total 6,000 Grass Carp (*Ctenopharyndodon idella*) (initial wt.=  $70.00 \pm 2.0$ ). The trail period is from August till November, 2020.Samples of five fish were collected after the time interval of 30 days and 50 at the time of harvesting (30 November). After sampling the growth parameters (Total body length, Total body weight, Condition factor and HSI) length-weight relationship, Viscerosomatic index (VSI %) and lipid content were analyzed. Total body length (cm) of Grass Crap increased from  $25.20 \pm 1.2 - 30.62 \pm 0.98$  cm, Total body weight (g) increased from  $502.0 \pm 117.54 - 806.67 \pm 63.79$  g, Condition factor increased from  $2.14 \pm 0.18 - 3.00 \pm 0.30$  %, Hepatosomatic index increased from  $1.59 \pm 0.14 - 2.58 \pm 0.24$  %. Viscerosomatic index was observed as  $6.06 \pm 0.23 - 13.43 \pm 0.23$  %. Regression coefficient (R²) value of Length-weight relationship was 0.534 which showed positive relationship between them. In conclusion, high stocking density in IPRS did not affect the growth in Grass Carp.

#### PREVALENCE OF METAZOAN PARASITES OF FRESHWATER TIRE-TRACK EEL MASTACEMBELUS ARMATUS FROM DISTRICT SANGHAR, SINDH PAKISTAN

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To determine the prevalence of metazoan parasites of freshwater Tire-Track eel *Mastacembelus armatus*, a total of 35 hosts were collected from the district Sanghar and brought to the Parasitology laboratory of Department of Zoology, University of Sindh, Jamshoro. Gut contents revealed the prevalence (82.86%) of metazoans in host fishes. Among them, the maximum prevalence (45.95%) was recorded for the Ergasilus, followed by cestodes (40.54%) and nematodes (13.51%). None of the host was found harboring trematodes and acanthocephalan.

# EFFECT OF HIGH STOCKING DENSITY ON *LABEO ROHITA* NUTRITIONAL QUALITY MAINTAINED THROUGH IN-POND RACEWAY SYSTEM

#### Qandeel Minahal, Shafaq Fatima, Wajeeha Komal, Shumaila Munir and Razia Liaqat

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In-pond raceway system (IPRS) is one of the sustainable and modern technology adopted in different countries that effectively increase fish production fulfilling dietary protein requirement. It was introduced in Pakistan for the first time in 2019. A total of 8,500 fingerlings of Labeo rohita (250.00  $\pm$  1.20 g) were stocked in each raceway in a water volume of 220 m<sup>3</sup>. For comparative study, 3,500 fingerlings (250.00 ± 1.20 g) were also stocked in each earthen pond with a land area of 6167m<sup>3</sup>. Stocking was done in replicates both in raceways and earthen pond. Fish samples were collected from both groups on monthly basis from June till November, 2019 to study the difference in fish nutritional quality reared at two different densities. Fish growth rate increased in both raceways and earthen ponds i.e.,1070 and 1090 g respectively at the time of harvesting. Feed conversion ratio was 1.90 in both groups. Mean survival rate was 99 % and no sign of any disease was observed in both groups. Fish nutritional quality was in desirable range in raceways. Crude protein in fish muscle ranged from 20.91±1.29 to 21.60 ±1.34 and there was no significant reduction as biomass was high. Fat contents was up to nutritional requirements of an adult human. High levels of polyunsaturated fatty acids (n-3 and n-6) such as eicosadienoic acid and decosahexanoic acid in fish muscles show that muscle quality is improved despite the high stocking density. Ten essential amino acids were determined among which lysine and leucine were most abundant. Among nine non-essential amino acids, glutamic acid was in high proportion. The results prove the efficiency of IPRS with improved production, growth rate, FCR, survival rates and nutritional quality of fish muscle.

# STUDY OF PROFILE OF FATTY ACIDS AND PROXIMATE COMPOSITION IN *TILAPIA* (OREOCHROMIS NILOTICUS) BY USING IN-POND RACEWAY SYSTEM

#### Wajeeha Komal, Shafaq Fatima, Qandeel Minahal, Shumaila Munir and Razia Liaqat

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Fish production would be enhanced in order to meet desired protein requirement. Production can be improved by adopting modern technologies. In- pond raceway system (IPRS) is the most modern and innovative technology in which high biomass would be reared to gain high yield. In 2019 this sustainable technology was introduced in Pakistan. Tilapia (*Oreochromis niloticus*) were stocked in raceways in replicates (initial weight=  $32.00 \pm 1.20$  g) at high stocking density (16,500) in 220 m³ water area. Fingerlings were stocked in earthern pond were taken as control group in replicates at stocking density of 4,500 in 6167 m³ land area. Monthly sampling was done from June till November, 2019 from both raceways and control groups to examine the proximate composition and fatty acid profile in fish muscles. There was increased in the growth rate of fish in both raceways and control groups  $769.50 \pm 51.43$  g and  $787.50 \pm 42.86$  g, respectively. Feed conversion ratio measured in raceways and control groups is 1.25 and 1.24, respectively. No sign of disease was observed in both groups. Mortality rate in both groups is less than 1%. The level of crude protein was in the desire range of  $(18.15\pm1.34 - 20.00\pm1.34\%)$ . The lipid content in fish muscles were also up to the desired range of 15.08 - 15.88 g in raceways and 16.00 - 17.20 g in control groups. High level of polyunsaturated acids (n-3 and n-6) especially eicosadienoic acid and decosahexanoic acid were found in raceways stocked under high density. It is concluded that IPRS is sustainable technology to obtain high yield from small area as the quality of fish did not alter due to high biomass.

# STUDY OF PROFILE OF FATTY ACIDS AND PROXIMATE COMPOSITION IN *TILAPIA* (*OREOCHROMIS NILOTICUS*) BY USING IN-POND RACEWAY SYSTEM

#### Wajeeha Komal, Shafaq Fatima, Qandeel Minahal, Shumaila Munir and Razia Liaqat

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# STUDY OF GROWTH PARAMETERS AND AMINO ACID PROFILE OF *TILAPIA* (*OREOCHROMIS NILOTICUS*) CULTURED IN IN-POND RACEWAYS SYSTEM

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In-pond raceway system (IPRS) is an innovative technology for sustainable aquaculture production by effectively increasing production, reducing pollution, improving feed and pond management. Fingerlings of Tilapia (*Oreochromis niloticus*) (n = 16,500, initial weight =32.00  $\pm$  1.26 g) were stocked in two raceways. Fingerlings (n = 3000, initial weight = 32.00  $\pm$  1.26 g) were also stocked in two earthen ponds to be studied as control. Average final weight achieved in raceways and control groups was 769.50  $\pm$  51.43 g and 787.50  $\pm$  42.86 g, respectively. In raceways, higher values of condition factor (2.80 %) were noted in June and November. Average feed conversion ratio observed in raceways and control groups were 1.25 % and 1.24 %, respectively. The highest value of specific growth rate was observed in June and it remained within the range of 0.30  $\pm$  0.14–0.68  $\pm$  0.02% over the study period. A significant difference (P < 0.05) was noted between amino acid profile in control and raceways. Highest concentration of leucine, lysine, threonine and arginine were observed in IPRS. Among non-essential amino acids were determined to be within the range of 29.72  $\pm$  0.25–32.23  $\pm$  0.1 mg/gcp in both groups over the study period. Total non-essential amino acids were noted be higher than total essential amino acids throughout the study period. Overall survival rate of tilapia was observed 99 % in both groups. It can be suggested that negative effects of high stocking density were not observed in tilapia when cultured, using IPRS technology.

# PREPARATION OF LOW COST FISH FEED BY PARTIAL REPLACEMENT OF FISH MEAL WITH COPRA MEAL FOR IMPROVEMENT OF CATLA CATLA FINGERLING'S PERFORMANCE

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Different types of plant-based alternatives were used to cope with high cost and availability of fish meal in aquaculture industry. Present experimental work was done to determine the effects of low cost feed prepared by using coconut meal based diets on overall performance of *Catla catla* fingerlings. Six experimental diets (0, 10, 20, 30, 40 and 50% replacement of fish meal) were prepared using copra meal as an alternative feed ingredient. Fingerlings were fed twice a day at 4% of their live wet body weight and feces from each tank were collected and were stored for chemical analysis for estimation of nutrient digestibility and mineral absorption. According to results it was observed that, coconut meal can replace fish meal from 10-20% and showed highest performance. Maximum nutrient digestibility (crude protein; 69.29%, crude fat; 69.14% and gross energy; 67 kcal/g) and mineral absorption (Ca; 74%, Na; 71%, K; 75%, P; 76% and Fe; 77%) was found in fish fed at 10% replacement of fish meal by following 20% replacement. Further increase in replacement levels resulted in poor digestibility in fish. So, it was cleared form the results that we can add copra meal at 10-20% in feed by replacing fish meal for making a cost effective and environment friendly fish feed.

# PARTIAL REPLACEMENT OF FISH MEAL WITH LOCALLY AVAILABLE BLACK SEEDS (NIGELLA SATIVA) FOR ROHU (LABEO ROHITA) FINGERLINGS

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The effect of black seed meal-based diet was investigated on the growth performance, nutrient digestibility, mineral absorption and hematological parameters of *Labeo rohita* fingerlings over the time period of ninety days. Six experimental diets were formulated by using black seed meal (0, 10, 20, 30, 40 & 50% by replacing fish meal) and feed pellets were formed. Fingerlings were given their respective diets two times in 24 hours at 4% level of body weight and feces sample was taken and preserved. According to results of present research, it was revealed that rohu fingerlings showed significant improvement when black seed meal was incorporated in fish diet at the level of 10 and 20%. Maximum weight gain (17.21g), weight gain percent (241%), specific growth rate (1.36) and best feed conversion ratio (1.31) were observed in fish fed on 20% of BSM. In the same way, highest digestibility of nutrients (crude fat, 72%; crude protein, 73% & gross energy, 69.51 kcal/g) and hematological parameters i.e. RBC (2.72×10<sup>6</sup>mm<sup>-3</sup>), Ht (35%) and Hb (8.10g/100ml) had their maximum values at 20% of BSM. Maximum level of mineral absorption of K (75%), Ca (71%), P (73%) and Na (73%) were also recorded at experimental diet III (20% of BSM). On the basis of these results, it was concluded that maximum of nutrients and minerals were absorbed in fish body at 20% of BSM improving growth and overall performance of fingerlings by decreasing discharge into water that will ultimately decrease water pollution.

# A PRELIMINARY STUDY ON ICHTHYOFAUNAL DIVERSITY OF MIR KALAM DAM, NORTH WAZIRISTAN

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In the present study, Mir Kalam Dam in North Waziristan was explored for the first time to identify its ichthyodiversity. The study lasted for nine months, from February to October 2018 during which a total of 550 fish specimens were collected and identified as 11 species, belonging to three orders, three families, and nine genera. Among them Family Cyprinidae was the most speciose and the richest Family, represented by nine species i.e. Cyprinus carpio, Carassius auratus, Crossocheilus diplocheilus, Tor macrolepis, Puntius waageni, Aspidoparia morar, Barillius pakistanicus, Barillius vagra and Barillius bendelisis. The remaining two families were represented by only one species each. Family Siluridae was represented by Ompak pabda and Family Chandidae by Chanda nama. The most abundant fish in the Dam was Cyprinus carpio (26.18%) while the least abundant fish species were Tor marcolepis and Barillius bendelisis (0.36%) each. Results show that 27% species found are edible whereas the rest of the small fish species are important members of the food chains present in the dam. Hence it can be concluded that the water of Mir Kalam Dam is productive and new species might be introduced in the Dam to provide economic support to the people of the area. Moreover, it is recommended that awareness projects should be conducted in North Waziristan for the ichthyo education in local people.

#### DEVELOPMENT AND SENSORY EVALUATION OF READY TO COOK FISH CRACKERS MADE FROM LABEO ROHITA MEAT AND DIFFERENT STARCH SOURCES

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Fish crackers are foods which contain comminuted fish and are classified as "half products" or "intermediate products". Most of the snacks available in the market are mainly based on cereals, which are high in calorie and low in protein content. For this reason, snacks like fish crackers with high protein content was thought to be developed for nutritional enrichment especially for chlidren. Different starch flours like rice, sago and tapioca, as a functional ingredient, were used to prepare fish cracker at 65:35 % ratio. The crackers (rice - C1, tapioca - C2 and sago C-3) were subjected to analyses for proximate as well as comparative physicochemical and sensory evaluation so as to determine the nutritive value and its quality attributes for general acceptance. Dried cracker had a moisture content of 9 to 11%, protein content of 20 to 25% whereas, lipid content found low but it significantly increased after frying due to oil absorption. Nutritionally all the treatments were at par with each other. However, maximum linear expansion was observed in tapioca based fish crackers was observed highest. Water absorption index (WAI) decreased while water solubility index (WSI) increased for tapioca based fish cracker. Maximum lightness and lower redness as well as yellowness was noticed with sago based fish cracker after frying. Fish crackers incorporated with tapioca starch had a maximum score for the sensory aspect of crispiness (4.7), texture (4.6), odour (3.8) taste (4.0) and overall acceptability (4.2). Storage tests showed increase in peroxide and flavonoid values and decrease in phenolic and antioxidant contents within prolong storage period.

# DEVELOPMENT OF INOVATIVE READY TO COOK AND READY TO EAT *LABEO ROHITA* SAUSAGES AND EFFECTS OF PHYSIOCHEMICAL SENSORY AND STORAGE PROPERTIES

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This study investigated sensory, proximate and storage properties of fish sausage made by three different cooking methods. Ready to cook and ready to eat fish sausages were developed using different cooking methods like boiling, smoking and frying. Fish mince (1Kg) was mixed with different spices by mixing crushed ice to form emulsified mixture. Mixed ingredients were placed and packed in casing and boiled at 100°C for 90 minutes, cooled in crushed ice, casing and packing was done. Fresh sausage was boiled and grouped as T1. T2 group include fried sausages and were shallow fried at 100°C for 10 minutes. Smoked sausage was smoked for 30 minutes at 75°C (T<sub>3</sub>). The sensory, proximate, physical quality and storage properties of these groups were compared with each other. Sensory evaluation showed best result for fried sausage than boiled and smoked. Proximate analysis showed high protein in smoked group while high fat in fried sausage than others. The storage parameter pH, antioxidant activity, flavonoid content and phenolic content of smoked sausages were high that indicate the long shelf life of smoked sausage than boiled and fried. Fried sausage had greater amount of fat that result in higher peroxide value that increase the oxidation of food. So, present study conducted showed that smoked sausage had long shelf life and aroma than fried and boiled sausage due to the presence of phenolic compound that maintain the shelf life of food. Sausage produced by different cooking methods are stored at 0, 10 and 20 days showed that the quality of fish sausage was not affected. Protein enriched ready to cook and ready to eat sausages can be beneficial for improving protein deficiency.

# ASSESSMENT OF MOLASSES SUPPLEMENTED FEED EFFECTS ON THE GROWTH AND SURVIVAL RATE OF INDIAN MAJOR CARP (*LABEO ROHITA*)

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The melting of sugar cane or sugar beets into sugar produced a sticky dark by-product which is called molasses. Molasses can serve as an outstanding source of fiber and minerals, it can be cost effective ingredient for animal feed. Moreover, molasses can enhance animal feed consumption and palatability. Fish is an excellent source of animal protein and good fat, its demand is increasing day by day. Currently, a lot of research had been done on fish feed additives that helped in increasing the fish growth and production. *Labio rohita* (fingerlings) is highly nutritious and commercial demanding fish in Pakistan. Therefore, the current research study was planned in order to assess the impact of molasses supplemented feed on the growth and survival rate of *Labeo rohita*. In the experiment, four kind of feeds were formulated by mixing 0, 3, 6 and 9% molasses/100g commercially available fish feed. After 28 days length gain (TLG), net body weight (NWG), feed conversion ratio (FCR), specific growth rate (SGR), net weight gain percentage (NWG), protein efficiency ratio (PER) and feed intake were examined / recorded. The results revealed that fish treated with 3% molasses/100g fish feed shown significant increase in TLG and NWG, as compared to control and other formulated diets. In addition, other growth parameters were also showed significant improvement with the same dosage feed. It could be concluded that molasses serves as potential feed supplement in fish feed to improve its growth and production but it could only be possible with more research on the composition of molasses and different concentrations in fish feed for different species.

# SPAWNING PERIODICITY OF CATFISH, *OMPOK PABDA* (HAMILTON- 1822) FROM RIVER INDUS, SINDH; PAKISTAN

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The spawning periodicity of *Ompok pabda* from Indus River was elucidated on monthly basis for collection during February 2017 to January 2018. The measurement of egg size, somatic index of gonads and productive potential (fecundity) was considered of the fish under study. Size of the egg was found increasing from March (0.81 mm) to May (1.05 mm) with one spawning peak in May. Somatic index percentage of gonads in both sex also noted increasing simultaneously (0.95% in March and (1.2% in May) for male and (3.18% in March) and (5.1% in May) for female. The value of both factors reflects that the fish possess only one spawning season during the year in the month of May. The enumeration of fecundity was based upon ten mature fish ranging from 15.5 to 30.0 cm in length and from 18.18 to 148.0g in weight. High potential of eggs production i.e. (1020 eggs) was recorded from fish with 30.0cm and 148.0 g in length and weight. The low fecundity (300 eggs) was counted from fish of 15.5cm in length and 18.8g in weight. Production potential (fecundity) and its association with other factors like total length of fish, weight of fish, weight and length of gonad were enumerated and the fecundity found to be dependent with gonad weight as compared to other factors.

# INCREASING THE DIGESTIBILITY OF CORNCOB AS FEED INGREDIENT BY UREA TREATMENT FOR GRASS CARP (CTENOPHARYNGODON IDELLA)

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The present study was conducted to determine the effect of corn cob treated with different levels of urea as feed for Ctenopharyngodon idella fingerlings. Four experimental feeds were prepared by treating corncob with graded levels of urea 0%, 2%, 4% and 6% respectively. Grass carp (Ctenopharyngodon Idella) fingerlings were fed with urea treated corncob containing for 90 days at 3% body weight. The fingerlings were divided into four treatment groups (T0, T1, T2, T3) each treatment having three replicates. After the completion of feeding trial growth parameters were analyzed and significantly increased final weight, weight gain, weight gain % was observed in T1 (2% Urea) than other treatments. Feed intake was improved by treating corncob with urea and significant maximum feed intake was seen in as T1 (45.04) was statistically significant (P<0.05) as compared to other treated groups. SGR and FCR showed significant (P<0.05) difference in the groups and better values were observed in T1 (2% Urea). Survival rate remained same in the experimental groups and showed non-significant (P>0.05) difference. Whole body proximate composition of Grass carp showed significant (P<0.05) difference as crude protein and dry matter and ash was high in T1. Hematological analysis showed significant (P<0.05) results of Hb, HCT and WBC, T1 showed higher Hb value. While RBC showed statistically non-significant (p>0.05) results. Nutrient digestibility improved by treating corncob with urea as better results were found in treated groups as compared to the control group moreover significantly (P<0.05) higher ADC (protein) and ADC (fat) was found in T1 (2% Urea) except ADC (dry matter). Intestinal digestive enzyme activity of amylase increased in the treated groups and showed statistically significant (p<0.05) results. Lipase and protease showed non-significant (p>0.05) results in control and urea treated groups. Physico-chemical parameters of water remained same throughout the experiment and showed statistically non-significant (p<0.05) difference. It can be concluded that urea treated corncob can be used as effective feed ingredient in the feed for grass carp.

# DIVERSITY AND DISTRIBUTION OF PONY FISHES (TELEOSTEI: PERCIFORMES LEIOGNATHIDAE) IN THE WATERS OF SONMIANI'S BAY BALOCHISTAN

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Leiognathidae species are commonly known as pony fish or slip mouths, the name derived from their highly protractible mouth, which protracts either dorso-rostrally, rostrally, or ventro-rostrally. They are small bioluminescent, schooling fishes, commonly found near-shore and estuarine in Indo-Pacific waters. They are mostly silvery in colour, size is generally small, and have compressed body. Eye moderate to large, preceded by a short, sub nosed snout. Leiognathidae represented with total 10 generas and 51 species. Among the ten generas of pony fishes (Perciformes: Leignathidae) Gazza Ruppell, 1835 is the only genus characterized that is having canine teeth anteriorly in jaw. The present study was carried out for one year, from October 2020 to September 2021 in Sonmiani bay Balochistan. These fishes were caught using gill net and beach seine during monthly regular collection. The seasonal variation were observed in the diversity and abundance through the year The most abundant species such as (*Photopectoralis aureus, Nuchequula blochii, Aurigequula fasciata, Photopectoralis bindus* and *Nuchequula gerreoides*) were found during pre-monsoon while least number of species were studied during North East monsoon that were *Leiognathus equula* and *Nuchequula gerreoides*. *Secutor interruptus* was the most abundant species as collected during high tide of southwest season whereas, the *Nuchequula gerreoides* was the only species found in high abundance during the three season except NE monsoon.

### 2. MARINE BIOLOGY

# DIVERSITY, ABUNDANCE, ZONAL DISTRIBUTION AND SEASONAL OCCURRENCE OF INTERTIDAL MACRO-FAUNA AT ORMARA WEST BAY, BALOCHISTAN

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The seasonal variation in diversity, distribution and abundance of intertidal macrofauna was studied in relation to physico-chemical parameters of water at sandy intertidal beach of Ormara West Bay, Balochistan. Samples were collected quarterly through quadrate methods during Autumn Inter Monsoon (AIM), North-East Monsoon (NEM), Spring Inter Monsoon (SIM), and Southwest Monsoon (SWM) seasons from two zones i.e. Low Tidal zone (LTZ; minimum exposed area) and High Tidal Zone (HTZ; maximum exposed area). In the present study 81 species were recorded and identified in five groups, i.e. Gastropods (41 spp), Bivalves (29 spp), Polychaeta (8 spp), Cnidarians (1 sp.), Scaphopoda (1 sp.), and Echinodermata (1 sp). Among these, Gastropoda and Bivalvia were the most dominant groups. A total of 41 species of gastropods belonging to 25 genera and 23 families were recorded from the high and low tidal zone. At both tidal zone genus Strioterebrum dominated during SIM followed by genus Bullia (SWM at low tidal zone) and genus Oliva in (AIM) and Cerithium (NEM) at high tidal zone. The 29 bivalve species, reported here from low and high tidal zones, were distributed in 15 genera and 11 families. The genus Tellina was most dominating during all seasons and tidal levels. Among all species, Tellina arsinoesis was the most abundant species followed by Vepricardium asiaticus, Mactra aequisulcata, and Didimacar tenebrica. The highest value for gastropods diversity, for example, Shannon Index was observed during NEM season, whereas Simpson's diversity was higher during NEM and AIM at both LTZ and HTZ. Distinct seasonal variation in air and water temperatures was recorded with the highest temperature during SWM and lowest during NEM, respectively reverse is true in the case of salinity. The maximum values of pH were observed in SIM. The highest concentration of DO was noted during SWM and lowest during NEM seasons. The concentration of nutrient ions appeared to fluctuate seasonally i.e. ammonium ions had the highest value during SIM, nitrate and nitrite ions during SWM and phosphate ions during AIM. The present study provides reference point on seasonal variation in macrofaunal diversity with relation to physicochemical parameters of water for the first time from an exposed sandy beach at Ormara West Bay, Balochistan. The species composition and abundance showed the significant impact of monsoon on macrofaunal diversity and abundance at studied beaches.

# SEASONAL VARIATION IN COMMUNITY COMPOSITION OF MESO-ZOOPLANKTON ASSOCIATED WITH MARINE SPONGE *LIOSINA PARADOXA* THIELE, 1899 AT MANGROVE FOREST KARACHI, PAKISTAN

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Marine sponge harbors highly diversified communities in mangrove forest of tropical and sub-tropical regions. Liosina paradoxa is the only species of marine sponge which was reported recently for the first time from mangrove forest at Karachi, Pakistan. The sponge samples were taken from pneumatophore of Avicennia marina. These sponges were collected from four transects during pre-monsoon, monsoon and post monsoon period. In this investigation meso-zooplankton community associated with sponge (Liosina paradoxa) was assessed. A total of 29 species (840 individuals) of six taxonomic assemblages included dominating community of Nematoda (40.60%)

followed by Crustacea (37.74) and Polychaeta (19.64%) whereas, other minor groups as Platyhelminthes (1.3%), Hemichordata (0.48%) and Rotifera (0.24%) were found with lower diversity. Among nematodes, the most abundant species was *Paracanthonchus* sp. followed by *Paracyatholaimus* sp. and *Eleutherolaimus inglisi*. Crustaceans was the second large abundant group including *Oithona* sp. and cypris larvae of *Balanus* sp. found in all transects. Among polychaetes, *Sphaerosyllis* sp. was the most dominant species in all transects. In case of minor groups, *Lehardyia* sp. (Platyhelminthes) was dominating and found in all transects while rotifers and hemichordates found in less number. According to diversity indices, species richness (Margalef and Menhinick) and Shannon-Weiner index was maximum during post monsoon (R1 = 0.535; R2 = 0.242; H' = 1.140, respectively), while Simpson index was highest during monsoon ( $\Delta = 0.665$ ). Dominance and evenness was observed maximum during pre-monsoon (D = 0.374) and post monsoon ( $\Delta = 0.782$ ), respectively. Further studies are needed to observe and understand the dynamics of meso-zooplankton inhabitant of different niches at mangrove forest along Pakistan coast.

# COMPARATIVE ANALYSIS OF OTOLITH MORPHO CHARACTERS FOR EIGHT SPECIES OF CLUPEIDAE FOUND IN DIFFERENT COASTAL WATERS OF PAKISTAN

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The polycarbonate structures of Otoliths of teleost fishes are composed mainly of aragonite, present in the inner ear in three pairs called sagitta, asteriscus and lapillus. In general, the otolith size and shape differ among species, among populations and within each species. These variations are influenced during development by both genetic and environmental factors and explore taxonomic variations. Objective of this study was to establish the identification features of otolith description and provide an ease to differentiate in closely resembling species. Eight species (Nematolosa nasus, Nematolosa Arabica, Hilsa Keele, Tenulosa ilisha, Sardinella longiceps, Anodontostoma chacunda, Sardinella melastoma, Tenulosa toil and Sardinella melastoma) of clupeidae were examined as otolith shape descriptions and morphometric (length, height, and weight) comparison from Sindh and Balochistan coast of Pakistan. The ontogenetic differences in otolith shapes have been analyzed through multiple  $\chi^2$  tests (significance 0.05) and it revealed morphological variations in rostrum, marginal structures, features of crista and Otolith length (OL): otolith height (OH), arm length (AL): Rostrum length (RL) ratios. Overall, this study shows that these morphological features of otolith can be used in the discrimination of clupeid species.

# IDENTIFICATION AND MORPHOMETRY OF STRIATOBALANUS TENUIS (BALANOIDEA) COLLECTED FROM INTERTIDAL ZONE OF ROCKY SHORE KARACHI

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The present study was designed to identify the morphometric relationship of species *striatobalanus tenusis* and to quantify traits of evolutionary significance by detecting changes in shape and function or evolutionary relationship. The specimens of *Striatobalanus tenuis* were collected from Sandspit and Buleiji. *S. tenusis* is deep sea specie (300-400 m) which is found from different gastropods shells like *Murex, Neverita didyma*. This species has been identified on the basis of size shape and number of calcareous plates surrounded by its body. The shell composed of six plates, conical, white orfice deeply toothed. Scutum triangular strongly sculptured on dorsal surface, Tergum, triangular with long and narrow spur. For morphometric relationship size among species has been expressed in terms of basal diameter of shell was measured, which was variable in response of width of carinal plates. The minimum size of basal disc was 0.9 cm and the maximum size was 1.9 cm in the collected samples. This parameter has continued to be used as an estimate of size. The length and width of carinal and rostral plates has also been measured. These morphometric measurements revealed the significant differentiation and accurate measurements between species.

# PROTEIN VARIATION THROUGH SDS PAGE ANALYSES AMONG THE TWO SPECIES OF BRITTLE STAR (*ECINODERMATA: OPHIUROIDEA*) FOUND ALONG THE COAST OF PAKISTAN

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The class Ophiuroidea is one of the largest group of individuals among the phylum Echinodermata with 2,096 recognized species. Morphologically, the typical body plan is composed of a pentagonal to round central disk and five arms that sharply head off from the disk. However, some species exhibit six, seven, or up to ten arms which can branch once or many times. The electrophoresis of enzymes on starch or polyacrylamide gels provides a powerful test of the validity of presumed species and also some preliminary estimation of protein of different weight. The two species (*Ophioneries dubia* and *Macrophiothrix aspidota*) of brittle stars were selected for the initial screening of general proteins, with known weight marker, using Sodium dodecyl sulfate (SDS) PAGE with Commassie blue stain. The specimens were collected from the Buleji coastal area. The individual species were varied according to disc diameter and arm size as disc diameter size ranged from 3 mm to 15 mm for *Ophioneries dubia* and 5mm to 24 mm for *Macrophiothrix aspidota*. The relative mobility of variable number of bands was calculated for both the species. The use of SDS PAGE vertical electrophoresis *provides the preliminary identification and discrimination of protein* on the bases of molecular weight. This selective basic data information will be helpful for further advanced studies.

# SOME PRELIMINARY OBSERVATION ABOUT THE PRESENCE OF MICROPLASTIC IN THE GUT CONTENT OF FOUR SPECIES OF MANGROVE CRABS

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Microplastic are tiny particles of plastic that produces by commercial product as well as breakdown of larger plastic molecules into smaller numerous and cannot be recycled and none can be recycled more than few times. This deadly fusion had made plastic global environmental adversity with catastrophic consequences for human and animals. The mangrove crabs are essential component of marine ecosystem and characterize a type of food that can be eaten easily by other sea food animals and humans Inspite of a current catastrophic issue no information is available from the gut content of crab species and microplastic accumulation in different tissues of crabs. In this study, the abundance and accumulation of microplastic in different tissues guts and gills of four species of mangrove crabs (*Deltuca urvillei, Metopograpsus thukuhar, Macrophthalmus pectinipes* and *Austruca iranica*) through overnight digestion process. After the digestion and filtration the sample was observed and observed through a stereomicroscope with charged coupled device (CCD) camera. The abundance of microplastic in the gut and gills of the different crab species was assessed and it was observed that the abundance of microplastic in the gut was significantly higher than the gills. Different morphotypes of microplastic were observed in crab samples mainly includes microbeads and fibers.

#### POPULATION GENETIC STRUCTURE OF MUD CRAB, SCYLLA OLIVACEA FROM PAKISTAN/NORTHERN ARABIAN SEA

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Large marine ecosystems (LMEs) are regions of the world's oceans, including coastal areas from river basins and estuaries to the borders of continental shelves. Productivity in LME protected areas is comparatively higher than the open ocean, the 66 LMEs produce about 80% of global annual marine fishery biomass. The Arabian Sea ranked

32 on a global measure of LMEs. *Scylla olivacea* (mud crab) is a significant reserve of aquaculture and commercial fishery in the (NAS) Northern Arabian Sea. The mt-DNA variations in *S. olivacea* (n14) were estimated from Sandspit back waters and Korangi creek mangrove areas populations from Pakistan. The present study based on Cytochrome Oxidase (COI) and 16S rRNA genes, high haplotype (*Hd* 1, *Hd* 0.978) and low nucleotide diversity (*PI* 0.04, *PI* 0.10) was observed in the populations of *S. olivacea* respectively. The neutrality tests (Fu's F'sand Tajima's) were non-significant, although mismatch analysis revealed the potential population expansion event occurred through Gene flow. Additionally a phylogeography analysis of *S. olivacea* based on the COI was estimated based on sequences obtained from GenBank (n99) fromIWP. Out of all sequences n113 (14 from Pakistan and 99 from Genbank), different COI haplotypes were identified. The AMOVA indicated the phylogeographic regional partition and genetic structure in IWP. In the current study, the partial sequences of the COI genes offerdirection with the evaluation of the genetic structure, phylogeography and genetic relationship of *S. olivacea* in the IWP region.

# SOME METRIC STUDIES IN FOUR SPECIES OF SEA ANEMONE (CNIDARIA: ANTHOZOA: ACTINIARIA) FROM MANORA COAST KARACHI, PAKISTAN

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Cnidaria are the sixth most diverse marine phylum in the world, where's the order Actiniaria commonly referred to as sea anemones a small, moderately diverse order, due to the intricacy of identification of sea anemones, this order has remained some unidentified species. Currently, over 1340 anemone species have been described throughout the world. Sea anemone inhabit on many organisms species i.e. fishes, sponges, algae, seaweed and provide food to many marine animals, they are highly successful in an ecological intelligence, an important part of the marine ecosystem. Four species of sea anemones (Cnidaria: Anthozoa: Actiniaria) were recorded during the survey of intertidal and subtidal rocky and sandy shores in Manora (Karachi, Pakistan). Identification was done on the basisof Morphological features and morphometric measurements. A total four species including *Bnudosoma sp. Diaduminatus lineatus, Actina aquina, Pseudectina flagilefera* were collected out of which three are previously reported and one is under taxonomic identification. Morphometric study from of 189 specimens of four species was performed in which column length, column width, diameter of oral disc was performed along with the density of an individual specie on known stratum, was also observed. The oral disc diameter was greater in *Pseudectina flagilefera* and was smallest in *Actina aquina*. Polyacrylamide gel electrophoresis was also performed to differentiate the species at molecular level and the result support the morphological findings.

# MOLECULAR AND SYSTEMATIC IDENTIFICATION OF NEW RECORD OF *METAPENAEOPSIS* STRIDULANS (DECAPODA: PENAEIDAE) ALCOCK, 1905 IN PAKISTANI WATERS

#### Syeda Hadiqa Noor\* and Noor Us Saher\*\*

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Within Penaeidae, *Metapenaeopsis* is the most diverse genus and *M. stridulans* is the only species which is confirmed from molecular and systematic identification. The traditional taxonomic identification revealed unsymmetrical petasma as a major identification tool while stridulating organ constituted a strongly supported feature. However, the morphological differences in *Metapenaeopsis* shrimps do not vary much, which make discrimination a bit tricky. In this study, the DNA barcoding and phylogenetic analyses used to examine taxonomy and phylogeny of genus *Metapenaeopsis* based on mitochondrial (COI) genes. In the present evolutionary analyses, which is done using MEGA 6 and the history were deduced using the Neighbor Joining (NJ) method. The phylogenetic neighbors based on the blast similarity sequences search between 88% plus. The evolutionary distances were calculated using the Maximum Likelihood method with the units of the number of base substitutions per site. The phylogenetic tree was constructed with the available sequences of Taiwan, China and Korea. Major clusters were

parted, where the main cluster clasping species which found near Taiwan and China. Taiwan's *Metapenaeopsis* showed much phylogenetic relatedness to the Pakistani species.

# SHELL ASSOCIATED CRABS (HERMIT CRAB), DISTINCTIVE MORPHOMETRY AND ITS PREFERENCE ALONG THE COAST OF PAKISTAN

#### Altaf Hussain Narejo and Noor Us Saher\*

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Paguridae are the most common families throughout the coastline of Pakistan. A total of 2149 individuals were collected from the intertidal water mark of three respective coasts i.e., Rocky, Muddy, and Sandy. Morphological and metric studies have been conducted between the hermit crabs and its host gastropod shell. Individuals were categorized according to size, sex and shell occupation. Crabs were measured according to Total length (TL), Cephalothoracic shield length (CSL), width (CSW), Chela length (CL), width (CW), and weight of crab with and without shell. Whereas, the shells were measured according to Total length (TL), width, aperture length (AL), width and shell weight (SW). For morphometric analysis; Total length of crab used as independent variable. Results reveal the significant relationship of total length between the two families; Diogenidae and Paguridae (P>0.05). Among 2149 hermit crabs, 93% individuals belonged to Diogenidae acquiring 93% whereas, Paguridae acquired only 6.9%. Total 26 families of shells found hosting the hermits, among them Muricidae, Turbinidae, and Certhiidae found dominant in the utilization by hermit crabs. The Sex ratio showed the dominance total males (N = 1390) with 64.68% dominated in respective coasts while female s(non-ovigerous = 667) with 31.03% and females (ovigerous = 92) with 4.28%. High number of used shell species utilization by smaller individual crabs indicated that occupation is influenced by the shell availability, while larger individuals demonstrate more specialized and particular occupation.

# COMPARATIVE MORPHOLOGY AND MORPHOMETRIC RELATIONSHIP BETWEEN OTOLITH AND BODY SIZE OF TWO COMMERCIAL SPECIES OF FAMILY SCIAENIDAE

#### Abdul Hameed and Noor Us Saher\*

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The genus Otolithes is a group of marine teleost fishes from the family Sciaenidae, which are commonly called croaker or drum fish and characterized by having two exposed canines in each jaw, from which it derives the common name 'tiger tooth croaker'. The species level identification of some species of Sciaenidae family is difficult due to their morphological similarities and they are identified by the study of morphological variation of otolith, gills and shape of swim bladder. This study was designed to observe and compare the body structure and otolith shape of two (*Otolithes ruber* and *Otolithes cuvieri*) commercial species of sciaenidae and the relationship between the otolith size and body size of these species. In present study it was found that the *Otolithes ruber* differ from the *Otolithes cuvieri*by having a shorter head depth, more gill raker numbers at the lower limb of the first gill arch, and more numerous swim-bladder appendage pairs. The Sagitta of both species was oval in shape, pointed downward but the otolith of *Otolithes ruber* is slightly tapered as compared to *Otolithes cuvieri*. The linear relationship between body size and weight of both species with their otolith size and mass shows strong correlation between fish length, otolith length and width, The Positive similarly (P<0.05) was observed between fish weight with Otolith mass and it was also concluded that the Otolith size directly related and depend on the fish size and fish weight.

#### MICROZOOPLANKTON AS INDICATOR OF WATER QUALITY IN MARINE ENVIRONMENT

#### Roomana Yasmeen, Nafisa Shoaib and Tayyaba Hamid

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Microzooplankton are heterotrophic and mixotrophic plankton, they play pivotal role in marine environment. They are the main primary consumers of phytoplankton. Planktons are used as bio indicator of pollution in marine environment, as they are susceptible to slight change in physical and chemical variation in marine ecosystem. Marine pollution due to anthropogenic activities and climate change induces negative effects on plankton abundance and biodiversity in marine ecosystem. Sea water samples were collected on board using Niskin bottles from the coastal waters of Pakistan monthly for a period of one year, abundance and diversity of microzooplankton were recorded employing standard methods. In our study 131 species of microzooplankton from the coast of Pakistan were identified. The highest abundance was recorded from station three which was near industrial area suggesting industrial activity affected microzooplankton. During the south west monsoon we examined the highest richness and abundance of microzooplankton in Baluchistan coast.

# ZOOPLANKTON COMPOSITION, DIVERSITY AND ABUNDANCE IN SANDSPIT, NORTHERN ARABIAN SEA

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Zooplanktons are heterotrophic plankton, range from 20 and 2000 µm in size, are weak swimmers and drift in water column of ocean, seas or fresh water bodies. The abundance of zooplankton and their biodiversity is linked to the health of marine ecosystems. The abundance and distribution of zooplankton are influenced by hydrographic condition and they have been suggested as good biological indicator species. Triplicate samples were collected monthly for a period of one year from Sandspit using zooplankton net, spatial distribution, abundance and diversity of zooplankton were recorded employing standard methods. The zooplankton abundance and diversity shows variations with different season and varied from near shore and offshore waters. The highest percent abundance recorded in Sandspit was of Bristle worm than Foraminifera and third abundance was of Calanoid Copepod Pollutants are known to reduce species diversity and increase population of tolerant species. The four seasons influence changes in physiological and chemical condition of marine waters determined species composition and distribution of zooplankton.

# DIVERSITY AND DISTRIBUTION OF NOCTILUCA SCINTILLANS (GREEN TIDE) IN THE COAST OF KARACHI

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The recent study based on annual distribution of Dinophycaea from Northern Arabian Sea, Karachi (Sandspit). Sea water was collected using Niskin water sampler from two stations for water quality and nutrients analysis. *Noctiluca scintillans* (dinoflagellate) is cosmopolitan in distribution and occurs in coastal environment. It is omnivorous, symbiotic and feed on planktons, eggs of fish and detritus. Maximum number of *Noctiluca* was observed 65360 cell/L in August than 10040 cell/L in December and 1620 cell/L in January. Among toxic species of dinoflagellates many genra were observed such as *Ceratium spp.* (12), *Dinophysis spp.* (3), *Gyrodinium spp.* (2), *Protoperidium spp.* (15), *Prorocentrum spp.* (6) and *Gonyaulax spp.* (4). The dinoflagellates are present as primary producers in marine water and produce bloom when population explosion occurs due to salinity, temperature,

excessive nutrients and anthropogenic activities. Some organisms produce toxins that affect filter feeding animals such as clams, oysters, mussels and crustaceans. Red tide caused fish mortality, affect fishery resources, biodiversity and threat to the marine environment. The increased frequency of algal blooms in the coastal areas worldwide is mainly due to human activities. As algal blooms cause hazardous impacts on coastal areas, economies and public health, so Government should develop strategies for controlling pollution.

### 3. PALAEONTOLOGY

# TRAGOPORTAX PUNJABICUS (ARTIODACTYLA: BOVIDAE) FROM THE DHOK PATHAN FORMATION OF MIDDLE SIWALIK. PAKISTAN

### Kiran Aftab 1\*, Muhammad Akbar Khan2, Mubashar Hussain1, and Areej Arif1

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New fossils of *Tragoportax punjabicus* have been recorded from the Dhok Pathan Formation, District Chakwal, Punjab, Pakistan. The *Tragoportax punjabicus* are medium-sized bovids having moderately hypsodont dentition with rugose enamel. The upper molars are quadrate in shape with less prominent styles and median ribs as compared to other genera of family Bovidae. The anterior median rib is stronger than the posterior rib. The entostyles are present on all molars. The *Tragoportax* originated during the middle Miocene and was heavily diversified during the late Miocene and finely got through extinction in the late Pliocene. Most of these species belong to the herbivore guild. This evidence proved that the Middle Siwalik environment was wet and humid with increase precipitation. So, their habitats were ranging from grassland to forests.

# NEW REMAINS OF *DORCATHERIUM* (TRAGULIDAE) FROM MIDDLE MIOCENE OF CHAKWAL, PAKISTAN

# Muhammad Asim<sup>1</sup>, M. Akbar Khan<sup>1</sup>, Muhammad Adeeb. Babar<sup>1</sup>, Khalid Meahmood<sup>1</sup> and Muhammad Akhtar<sup>2</sup>

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In current study, the new dental material collected from three sites of Chinji Formation, are described and discussed. The recovered material includes the cheek teeth including deciduous as well as permanent. The morphology and size of studied specimens here match with two *Dorcatherium* species *D. minus* and *D. majus*. The aim of study is to enhance knowledge on middle Miocene *Dorcatherium* specially to document the deciduous premolar of *Dorcatherium* which are least known.

# SIAMOTRAGULUS FROM THE CHINJI FORMATION OF THE CHABBAR SYEDAN, JHELUM, PUNJAB, PAKISTAN

Muhammad Akbar Khan<sup>1</sup>, Sayyed Ghyour Abbas<sup>1</sup>, Muhammad Khalil Nawaz<sup>1</sup>, Muhammad Adeeb Babar<sup>1</sup>, Muhammad Asim<sup>1</sup> and Muhammad Akhtar<sup>2</sup>

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Chabbar Syedan is a small village situated at the base of the Bakrala ridge, Jhelum, Punjab, Pakistan. The outcrops of this village represent the Chinji Formation by lithology and faunal elements. Field campaigns from 2016 to 2019 in these outcrops resulted in the collection of some good specimens including the artiodactyls and perissodactyls. Fossil

remains of family Tragulidae are also present. Though small in number but indicate the presence of genus *Siamotragulus*. *Siamotragulus* dental remains have not been described from the Siwaliks. Two specimens are extremely small in size and these may represent a new species. A detailed analysis of these specimens in progress.

# CHALICOTH ERE (CHALICOTHERIIDAE: PERISSODACTYLA) FROM POTWAR PLATEAU, NORTHERN PAKISTAN: RARE FINDINGS

Sayyed Ghyour Abbas<sup>1</sup>, Muhammad Akbar Khan<sup>1</sup>, Muhammad Adeeb Babar<sup>1</sup> and Asra Ghous<sup>1</sup>

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Chalicotheres are rare throughout the world and the same is applicable to these clawed herbivores in the Siwaliks. Eight new dental remains have been described from four localities of the Chinji Formation (14-11 Ma), Lower Siwaliks of Pakistan. The new specimens are attributed to *Anisodon salinus*, previously known as *Chalicotherium salinum*. The specimens are significant not only because of their rarity but also because of what they add to information about the morphological and metrical values of this species. The phylogenetic status of the Siwalik species is still debated and still requires a detailed analysis.

# TETRACONODON AND SIVACHOERUS (SUIDAE) FROM POTWAR PLATEAU, NORTHERN PAKISTAN

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Rare tetraconodonts, *Tetraconodon magnus* and *Sivachoerus prior* are ascribed in this article. *Tetraconodon* is believed to be occurred in the Upper Dhok Pathan and Tatrot formations of the Siwalik Group. However, the specimens described here from the mid Dhok Pathan Formation are important in relieving the long-held notion of the previous researchers. Similarly, *Sivachoerus prior* appeared earlier than it was thought previously.

## 4. WILDLIFE, DIVERSITY AND CONSERVATION

#### ECOLOGICAL CAUSES OF THE DECLINE OF WHITE-BACKED AND THE RED-HEADED VULTURES IN SINDH PROVINCE

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Five species of vultures in Sindh prefer to eat meat from the dead livestock. The White-backed Vulture, Redheaded Vulture and Long-billed Vulture are sedentary species, while the Eurasian Griffon Vulture and the Cinereous Vulture are winter visitors. The carcasses of the domestic livestock, which have residual drug 'Diclofenac' left in their meat, during the treatment of their sickness, cause mortality of these vultures, as this drug is toxic to them. It is still in use, despite the ban on its sale. These vultures are extinct in the province, except in the south-eastern areas. A declined population of White-backed Vulture, Red-headed Vulture and the Long-billed Vulture, still breeds in the south-eastern Sindh. The Long-billed Vulture is a cliff breeder and it breeds on the cliffs of Karoonjhar Hills, Nagarparkar. The White-backed Vultures and the Red-headed Vultures are tree breeders. These still survive in south-eastern Sindh, because of the nonavailability of veterinary treatment of sick livestock in the desert. Same is the case across the border fence, in India, where more cattle stray in the desert and are not treated, if sick. The dead animals in this area, had no Diclofenac shots and so, there is no mortality of vultures from this cause. The White-backed Vultures and the Redheaded Vultures do not get suitable breeding trees for the reason that a vast number of trees are with their branches lopped, on which vultures cannot make their nests or even roost for the night. An annually lopped tree remains useless for vulture nesting. Sometimes vultures have to make nests on fabricated high tension electric towers. Considerable number of trees are lopped to provide fodder for the goats and sheep herds. Prosopis cineraria is most abundant and preferred fodder tree in the area and the vultures commonly nest in it. The leaves and soft branches are also sold in the market as fodder, during the drought seasons. Trees have to be lopped, because the domestic livestock herds have overgrazed or over-browsed the Thar Desert Habitats. Vegetation palatability and quantity eaten by individual goats and cattle was experimentally determined. The carrying capacity of the habitats, where domestic livestock grazed or browsed was measured. Population estimates of the free grazing domestic livestock were made. It was also found that the livestock has to travel longer distances from their night-stay enclosures, to fill their stomachs in the degraded habitats of Thar desert. Less nutrition and long distance traveling in the mornings, evenings and during foraging, keep them hungry and weak. As a result, these fetch low price for the herder communities. Most herders purchase fodder trees seasonally for lopping. That gives a negative impact to the tree nesting vultures. The soft parts of the bodies of the livestock, such as ears and between the legs are infested with hard ticks. Their night roosting enclosures also had a number of soft ticks hidden in the sandy floor, which sucked their nutrition at night. These parasites not only further weaken the animals but also cause various diseases and thus cause more mortalities, bringing poverty for the herder communities. The herders never remove the hard ticks or clean sand from the night-enclosures. The awareness level of the communities is very poor. They remain illiterate. Their population growth is steep high. Each teenager male has to have a herd of livestock for grazing, to live. These interlinked factors, brought decline of the vultures. For the recovery of tree-nesting vultures, some suitable trees were purchased to save these from being lopped. The vultures started nesting on these trees. Thus, the ecology-based conservation measures worked.

# FOOD CONTENTS ANALYSIS OF WATERFOWLS PASSING THROUGH INDUS RIVER AT TAUNSA, SOUTH PUNJAB, PAKISTAN

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A study was conducted to investigate the food contents of migrating waterfowl in Bet Makwal Kalan near Head Taunsa Barrage, along the River Indus passage, South Punjab, Pakistan. A sum of 459 gizzard samples was collected, during their fall migration, relating to 10 waterfowl species i.e. Northern Pintail (n=56), Eurasian Wigeon (n=17), Garganay (n=07), Mallard (n=02), Northern Shoveler (n=22), Common Teal (n=129), Gadwall (n=158), Tufted Duck (n=04), Common Pochard (n=29) and Ferruginous Duck (n=34). The waterfowl species preferred to consume plants over animals as food during winter migration, while passing at Taunsa. Food analysis showed that all ten bird species ingested seeds abundantly. Two plant species (*Naias marina* and *Potamogeton pusillus*) were consistently among the most consumed seeds in six species of ducks including Eurasian Wigeon, Garganey, Mallard, Gadwall, Tufted Duck and Common Pochard, out of ten species. Whereas, the seeds of *Potamogeton pectinatus* and *Poligonum spp*. were found abundant in the diet of four ducks' species, i.e., Northern Pintail, Northern Shoveler, Common Teal, and Ferruginous Duck. In animals, mollusks and crustacean were well represented in gizzard samples. Additionally, study highlights the importance of vegetation in wetland ecosystem.

# GIS BASED EVALUATION OF POPULATION DENSITY AND DIVERSITY OF BIRDS OF PREY OF DISTRICT MUZAFFARGARH, PUNJAB PAKISTAN

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Diversity and estimation of populations of raptors were carried out in District Muzaffargarh (South Punjab, Pakistan) during January-March 2021, using belt transect (5 × 0.1Km) technique. We recorded 447 raptors, belonging to 10 species falling in 2 families (Accipitridae and Pandionidae) with a density of 34.38birds/Km². Values of dominance (0.185), Simpson 0.818, Shannon Wiener 2.00, and evenness (0.567) were calculated, suggesting an even distribution of the species Black Kite (*Milvus migrans*; relative abundance, RA 0.082) was most abundant species, flowed by Shikra (*Accipiter badius*; RA 0.038), Marsh Harrier (*Circus aeruginosus*; RA 0.0336), Black Shoulder Kite (*Elanus caeruleus*; RA 0.0537), Honey Buzzard (*Pernis ptilorhynchus*; RA 0.0201), and Brahminy's Kite (*Haliastur indus*; RA 0.0157). Other five species (Steppe Eagle, *Aquila nipalensis*; Common Buzzard, *Buteo buteo*; Long Legged Buzzard, *Buteo rufinus*; White Eyed Buzzard, *Butastur teesa*; and Osprey, *Pandion haliaetus*) were rare (RA 0.0022).

# FEEDING ECOLOGY, THREATS AND CONSERVATION MANAGEMENT OF KALIJ PHEASANT (LOPHURA LEUCOMELANOS) IN AZAD JAMMU AND KASHMIR, PAKISTAN

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Pheasants are bioindicators of our ecosystem and their population is declining. We investigated diet composition and threats to kalij pheasant Lophura leucomelanos to prepare conservation and management strategies in Azad Jammu and Kashmir, Pakistan. The diet analysis was done through crop contents of kalij pheasants, threats were assessed through field surveys, communities meeting, and data from 250 questionnaires during April 2020 and March 2021. Based on analysis of threats data, conservation management strategies are recommended. The diet analysis shows that mostly the kalij consumes plant matter as the major diet. We recovered 45 plant species in major, minor, and trace forms which consisted of seeds, leaves, flowers, fruits, rhizomes, and bulbs. Invertebrates including ants, insects, larvae, and grit were also recorded. According to respondents the highest sighting (62.4%) of kalij pheasant was recorded from the forest, followed by cultivated land (20.4%). Major threats to kalij pheasant include forest fire (41.6%), followed by hunting (27.2%), habitat destruction (18.8%), and natural predators (12.4%). The hunting (n=142) of kalij and hunting index (0.855) was recorded during the study period. The maximum hunting was in the evening (54.23%, n=77) followed by night (28.87%, n=41) and the main purpose was food. Stealing of eggs and chicks capturing was recorded from many sites. As per respondents, local community is also concerned about the conservation of this species. Development of more protected areas for conservation, awareness education, implementation of wildlife laws, patrolling of officials in the breeding season, and long-term monitoring plan can help in the conservation of kalij pheasant.

# MORPHOLOGY AND DISTRIBUTION OF *COLUMBA LIVIA* (GMELIN, 1789) AND *STREPTOPELIA DECAOCTAO* (FRIVALDSZKY, 1838) IN DISTRICT MIRPURKHAS, SINDH, PAKISTAN

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The Columba livia and Streptopelia decaoctao are the members of order columbiformes and family columbidae. They have very adaptive nature. These species are commonly seen everywhere. A study was proposed to determine the morphology and distribution of the C. livia (rock pigeon) and S. decaoctao (eurasian collared dove) in different areas of district Mirpurkhas as they are important for food value, educational value, pollination, agriculture value and ecosystem value. For the collection of data, field surveys were carried out from January, 2021 to July, 2021 for observing for the observation of species in question using binoculars and DSLR cameras. The morphological characteristics and variations were recorded and species were identified using identification keys and taxonomic literature. Morphological characteristics of C. livia were recorded as body coloration was dark grey, green purple shade on neck. Iris color was orange, red and dark brown, bill color was dark grayish while feet color was pink to red. The morphometry of C.livia was recorded as: body weight 316.8±39.7g (grams), body length 10.4±0.5in (inches), tail length 4.3±0.2in, wingspan length 13.2±0.6in, beak length 1.7±0.1cm (centimeter). S.decaoctao body was pinkish grey to buff grey. On nape a half collar ring was present in black with white edged. Iris color was deep red and pupil black. Bill color was black while feet color was dark purplish to brown. The morphometric parameters of S.decaoctao were recorded as followed: body weight 144.8±6.7g, body length 10.4±0.2in, tail length 4.6±0.1in, wingspan length 7.1±0.4in, beak length 1.4±0.1cm. Present study recorded the distribution of C.livia, in urban and suburban areas, whereas the populations of S. decaoctao were recorded from rural areas especially in agricultural habitats. Population density of C. livia was founded denser than population status of S. decaoctao.

# MORPHOMETRY AND CONSERVATION STATUS OF *ANAS PLATYRHYNCHOS* (LINNAEUS, 1758) OF HAMAL LAKE, SINDH, PAKISTAN

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Hamal Lake, one of Pakistan's largest freshwater lakes, is a wildlife sanctuary that provides shelter to a variety of not only resident but Siberian migratory birds such as ducks, flamingos, geese, coots, egrets, shorebirds, herons, cormorants, ibises as well. Present study was proposed to examine morphology and various threats that Anas platyrhynchos (mallard duck) face in the aquatic ecosystem of Hamal Lake. The taxonomic and ecologic status of A. platyrhynchos was ascertained through deep study of their morphological and environmental characteristics. Species was identified via morphology and literature review, while ecological conditions was analyzed thorough observations carried out during field surveys. Various threats such as habitat destruction, hunting, and anthropogenic encroachment were recorded. The morphometric of female specimens of A. platyrhynchos was recorded as: body weight (g) 999.5  $\pm$  163.9, Body length (in) 19.9  $\pm$  1.92, Tail length (in) 2.5  $\pm$  0.6, wingspan (in) 20.2  $\pm$  1.1, while morphometry of male specimens of A. platyrhynchos was recorded as: body weight (g)  $909.7 \pm 222.5$ , Body length (in)  $20.4 \pm 1.5$ , Tail length (in)  $2.6 \pm 0.5$ , wingspan (in)  $20.3 \pm 1.17$ . Minor morphological variation was record between male and female of species in question, whereas no any major morpho-taxonomic variation existed. The conservation status of A. platyrhynchos was observed very poor in the wildlife sanctuary as there was no implementation of conservation actions for saving wildlife. Hunting and poaching were observed common, meanwhile freshwater habitat was also recorded to be degraded through massive pollution created by local people. It was observed that a large number of ducks were captured from the Hamal lake for illegal trade that occurs openly in various markets of Qambar Shahdadkot.

# TAXONOMIC AND GENETIC RELATIONSHIP OF FIVE SPECIES OF CORVIDAE (AVES: PASSERIFORMES) BASED ON MITOCHONDRIAL COI GENE SEQUENCE FROM PAKISTAN

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The corvidae (Aves: Passeriformes) is a morphologically diverse family of birds. It is globally, well identified by DNA barcoding but in Pakistan their genetic identification and relationship estimation based on mitochondrial COI gene (cytochrome c oxidase subunit I) is under study. In current study, aim was to identify collected samples genetically and re-establish genetic relationship among them for this purpose five species of family corvidae (*Corvus splendens, Corvus corax, Corvus macrorhynchos Urocissa flavirostris* and, *Dendrocitta vagabunda*) were collected from different regions of Pakistan. Samples were barcoded and genetic relationship was established using different methods (Neighbour joining, Maximum Likelihood, Minimum Evolution, Maximum parsimony and UPGMA in MEGA7. The pairwise distance showed minimum value (0.041) between *Corvus splendens* and *Corvus macrorhynchos* and highest (0.165) between *Corvus splendens* and *Dendrocitta vagabonda* indicating less genetic distance among species of same genera and more among species of other genera with overall 0.122 average distance calculated at 1000 bootstrap repetitions. Reconstructed Phylogenetic trees has discriminated all five species into two distinct clades, one comprising Urocissa and Dendrocitta genus with one species each and the other with three species of corvus genus. This baseline study will provide data for geographic studies on birds in future. It has also proved barcoding as an effective molecular tool for species genetic identification and phylogenetic relationship inference.

#### ESTIMATION OF HEAVY METALS IN FEATHERS OF MIGRATORY WATERFOWL

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Samples of breast feathers were collected from three sites Chashma Barrage, Taunsa Barrage and Trimmu Barrage to estimate the concentrations of heavy metals (Cu, Ni, Cd, Fe & Zn). Breast feathers of two waterfowl species, Anascrecca (Common Teal) and Marecastrepera (Gadwall) were collected from hunters who granted from wildlife department to hunt migratory birds under special shooting permit. Samples were collected during two different time intervals October-November and March-April. Heating drying oven (IM-30) having an air circulation system was used to dry the breast feather samples at the temperature of 60°C for 48 h. Breast feathers show better indication of degree of metal exposure than other feathers and organs. Acid digestion of prepared samples was performed in the mixture of hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>) and warm nitric acid (HNO<sub>3</sub>) by using magnetic hot plate. Atomic Absorption Spectroscopy (AAS) was used to analyze the digested samples of breast feathers. Slightly difference was found in the heavy metal concentrations in the breast feather samples collected from three sites. Distribution pattern of these heavy metals was as: Fe > Zn > Cu > Ni > Cd in both species of waterfowl. Results of this study showed heavy metal contamination in the breast feathers of waterfowl. Higher level of metal concentrations indicated heavy metal contamination at three study sites in the Punjab Pakistan. Higher level of heavy metal contaminants might be due to few possible reasons such a local contamination source and accumulation of heavy metals in the diet of foraging waterfowl birds at different localities. Heavy metal concentrations are relatively serious and should be a cause of concern.

# HABITAT SELECTION OF COMMON MOORHEN (GALLINULA CHLOROPUS) DURING BREEDING IN IMPOUNDED MARSH WETLANDS IN DISTRICT BAHAWALPUR, PUNJAB

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This study has provided basic insight in to the habitat selection of common Moorhen during breeding in the area of Bahawalpur at different locations. First of all vegetation survey was conducted and the flora species of Acacia nilotica, Prosopis juliflora, Dalbergia sissoo, Typha elephantiana, Cynodon dactylon, Saccharum bengalensis, Solanum nigrum, Calotropis procera, Phoenix dactylifera and Zizyphus numularia were recorded which were most commonly present in the habitat of moorhen. Typha elephantiana was the most abundant and preferred habitat for moorhen during breeding season. Moorhen nests were searched through visiting all possible sites and monitoring the activities of the adults at dawn and dusk. Moorhen only used the microsites to build nests on the branches of trees or emergent vegetation bended on water up to 3-5 feet above the water level. These birds used foliage of Dalbergia sissoo, Prosopis juliflora and Acacia nilotica and other nearby vegetation as hide. Soil and water samples were also analyzed for selected habitat of moorhen. Data obtained is a major addition in existing meager information on common Moorhen, its habitat use, breeding biology preferred by this species in its habitat.

#### MORPHOMETRIC VARIATION OF SPINY TAILED LIZARD (SAARA HARDWICKII) FROM CHOLISTAN DESERT, PUNJAB, PAKISTAN

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This study has provided the data on morphometric of spiny tailed lizard (*Saara hardwickii*) in the area of lesser Cholistan desert, Punjab, Pakistan. Adult spiny tailed lizards were collected from poachers with the help of Punjab wildlife department, Pakistan. In Cholistan, these lizards are often illegally collected and sold in various parts of the country for their use in medicine and for making oil from its fat, for which poachers claim that; it is effective in joint pain relief and provides strength to the male sex organ. They were captured in the season of early winter. Among them 9 were male and 8 were female. Total body mass (g) were recorded (Female=142-182g to Male=200-371g). Total body length (BL) (range in male 160-200mm and female was 120-170mm) was also measured. Snout vent length (SVL) was measured for all spiny tailed lizards (range 120-150mm in male and 110-130mm in female). Tail length (TL) was noted (range 130-190mm in male and 140-160mm in female) and spines mark on the tail were also counted which were 25-31 in male and 21-25 in female. All measure was made by using digital LCD verneir caliper and weighing balance (SK-5KModel) having range of 1g to 5kg. Comparison between male and female morphometric was made using the Analysis of Variance (ANOVA). The *f*-ratio value is 21.4353. The *p*-value is .000327. The result is significant at p < .05. Detail study is required on ecology of spiny tailed lizard for its conservation in lesser Cholistan desert; one of main area of its distribution in Pakistan.

# BEHAVIOR AND FEEDING ACTIVITIES OF WILD ANIMALS IN BAHAWALPUR ZOO, PUNJAB, PAKISTAN

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Current study was conducted on behavior and feeding activities of wild animals present in Bahawalpur Zoo. The main objective of the study was to analyze the effect of captivity on the feeding and behavior activities of wild animals present at zoo. Activities like active, passive and abnormal were observed during the study. Observations were taken on morning and evening basis specially at the time on which diet was provided, amount of diet, food presentation and dispersal ways, animal's preference for food, induction of novel objects, presence of any feeding enrichment technique, hygienic conditions regarding food given and enclosures. Various stereotypic behaviors have been reported like self-aggression, head tossing and pacing. Such types of behavior were most commonly observed in Lion and Puma. Baldness and head tossing in one of the bears was observed that is the result of poor animal welfare. Unhygienic conditions in lion's enclosures were also observed. Food provided to the animals like Desert cat, Jackal, Asiatic cheetah, Indian wolf and Puma is less than the per day caloric requirement of the animals. Habitat management, feeding enrichment and the proper medications would be the best solutions to improve habituation under captivity. It will also increase the rate of reproduction in animals.

#### ABUNDANCE ESTIMATION AND FACTORS AFFECTING OCCURRENCE OF NARROW-MOUTHED FROGS IN SUBTROPICAL SCRUB FOREST, PAKISTAN

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The family Microhylidae (Gunter, 1858) is a well-defined monophyletic group of diminutive frogs. In Pakistan, the family is represented by two species: *Uperodon systoma* and *Microhyla nilphamariensis*. Although these two frog species are listed as Least Concern in the IUCN red list of threatened species, the former is recognized as rare in Pakistan. When a species is not detected at certain locations, it can indicate that the species may be truly absent or that it was not detected despite concerted efforts which could pose a potential serious issue for any conservation efforts (MacKenzie et al. 2002, Pollock et al. 2002). We carried out the present study to estimate abundance and to determine if the likelihood of occurrence of the Balloon Frog (*Uperodon systoma*) and Ant Frog (*Microhyla nilphamariensis*) was influenced by the site and survey covariates using binomial logistic regression in a subtropical scrub forest (Rawalpindi-Islamabad, Pakistan). Despite our best efforts, Balloon Frogs could not be detected from any site. A total of 37 individuals of Ant Frog were detected from the selected sites (33%). The Ant Frog was recorded from five of 16 (naïve occupancy 31%) selected sites of Islamabad Capital Territory and three of the eight sites (37%) of Rawalpindi Tehsil. The model yielded non-significant results for the Ant Frog, and correctly classified 87.5% of the cases (presence=75%, absence=93%). Our findings could be used to update the conservation status of the studied species.

## 5. BIODIVERSITY, ECOLOGY

# CO-RELATION OF HERPETOFAUNAL DIVERSITY WITH WILD FLORA OF NALLA CANE IN BAGH, AZAD KASHMIR

#### Abu ul Hassan Faiz, Sadaf Rubab, Muhammad Shahbaz and Lariab Zahra Faiz

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The population dynamic Herpetofauna fauna are indication of environmental pollution and also indicate the health of terrestrial ecosystem. The population of herpetofauna is under threat due to various habitat degradation elements. The present study is designed with objective to record base line data of herpeto fauna with their aquatic vegetation. The study was conducted from 20 January to 20 September 2019 in three areas (Shujabad, Gehal, and Mohri Ferman Shah) of district Bagh, Azad Jammu and Kashmir. The study records three species of amphibians (Himalayan toad, Hazara toad, Kashmir frog) and status of reported species are abundant while among serpentine fauna (Oriental rat snake, Buff striped keel back, Indian cobra, Himalayan Pit-viper, Caspian cobra). The reported Indian cobra, Caspian cobra are endangered species while rest of serpentine are abundant in status and distribution. Among lizards, three species are reported (Laudaka agrorensis, and Oriental garden lizard are abundantly distributed. The major type of vegetation was also reported (30 species of plants) in the sampled area. The vegetation was sampled through quadrate method and a total of 15 quadrates were taken in whole of study area. The PCA (principal component analysis) was applied to find correlation of habitat and species by Xl. State (version 4.1). The Himalayan toad, Hazara toad, Kashmir frog are in close habitat association while three species Indian cobra, Himalayan Pitviper, Caspian cobra, vary in habitat use according to results of PCA. The hierarchical clustering results 3 clusters on habitat similarity of amphibians, serpentine and lizards communities. The study provide baseline data for future research studies to find population dynamics of amphibians and reptiles.

# DIVERSITY ESTIMATION AND SPATIAL DISTRIBUTION PATTERN OF MAMMALIAN FAUNA IN MURREE KOTLI –SATTIAN KAHUTA NATIONAL PARK

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Biodiversity is distributed unevenly across spatial and temporal scales on earth. Knowledge about species diversity, distribution and activity pattern is essential for management and conservation actions. The study of species diversity is prerequisite to understand the health of an ecosystem. Presence/absence data obtained through latest rising and professional scientific technique is low-cost and their use is irritating. The current research was conducted to enlist the mammalian diversity of Murree Kotli Sattian Kahuta National Park, Pakistan, from 24 August 2020 to 30 July 2021. Camera traps were installed 19 locations within selected grids and remained active for 254 trap nights. Results obtained from primary and secondary data revealed the presence of 32 species of Mammals Viz, Barking Deer, Grey Goral, Leopard Cat, Jungle Cat, Kashmir Or Hill Fox (Vulpes Vulpes), Asian Palm Civet (Paradoxurus Hermaphroditus), Wild Boar (Sus Scrofa), Asiatic Jackal (Canis Aureus), Rhesus Monkey (Macaca Mulatta), Common Leopard (Panthera Pardus), Small Indian Civet (Viverricula Indica), Leopard Cat (Prionailurus Bengalensis) and Indian Crested Porcupine (Hystrix Indica). The highest photo-capture events were recorded for Kashmir or hill fox followed by leopard and Asiatic jackal. The species which frequently shared the habitat were Kashmir or hill fox and leopard, at same station moderately captured wild boar. The leopard cat, Asiatic jackal, rhesus monkey, small Indian civet and Asian palm civet intermittently shared the habitat. This will help to understand sympatric relationship and resource competition among various species. ArcGIS software was used to develop maps

of spatial distribution pattern of recorded mammalian species from the study area. We also investigated the human and mammalian funna interaction in the study area. Common leopard, Rhesus monkey, Wild boar, Indian crested porcupine, Bats species were identified as the possible problems of animals in the study area. Leopard and other predator's species like fox and jackals are responsible for the attacks on livestock and poultry while wild boar, rehashes monkey and porcupine were responsible for crop damages in the area, incidence of leopard attacks on local people were also reported in the study. Majority of local community is against the presence of problems animals in the study area. The study reports show great ecological potential and an immense challenge in the form of dependency, as well as diversity of the area in the form of species richness, and utilization of park resources by the native community. We recommend an extensive camera trapping to understand the habitat use and resource competition among the mammalian species of Murree KotliSattian Kahuta National Park.

# STATUS OF *CHELONIA MYDAS* (GREEN SEA TURTLE) AND HABITAT AT HAWKE'S BAY, KARACHI, SINDH, PAKISTAN

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Chelonia mydas (green sea turtle) is among the five turtle species that exist in Pakistan and it is listed as endangered species in IUCN red list. Considering their endangered status, present study was proposed to record current status of Chelonia mydas as well as its main habitat "Hawke's bay" at Karachi. Several surveys were carried out in between sunset to late night from August to November for observing the nesting condition of green sea turtles and rate of pollution along their habitat. The nesting behaviour was recorded during night only, however status of nests and habitat was observed during both day and night. Information about habitat of Chelonia mydas (green sea turtles) was also collected from the Turtle conservation unit. The results showed that green sea turtle faces wide variety of problems such as high rate of pollution, existence of large number of predators, human encroachment and disturbance at Hawke's bay Karachi. Feeding on plastic materials was observed as one of the main causes of their deaths. Sea turtle were observed to get choked to death due to consuming plastic bags and water bottle caps. Numerous nests of green sea turtles were found deteriorated by the predators like dogs, crows, skunks etc. Due to lack of conservation efforts, the green sea turtles were observed facing the loss of habitat and climate change as well. Green sea turtle was observed being affected by the large number of light plastic and other marine debris at Hawke's' bay. Besides that, sound pollution, thermal pollution and chemical pollution also threat to marine life as well the terrestrial life of turtle at the beach. Therefore, there is need of increasing the awareness among local communities about the importance of green sea turtle as they are easily susceptible to anthropogenic activities at every stage of their life. In this context, it is very important to implement strict conservation rules for the conservation of nests and the juveniles of green sea turtles. Implementing the conservation efforts and educating the local people are the serious urge for the conservation of green sea turtles at Hawke's bay.

# MORPHOLOGY AND DISTRIBUTION OF *TRERON PHOENICOPTERA* (YELLOW FOOTED GREEN PIGEON) IN SINDH, PAKISTAN

Abdul Rehman Shaikh, Kalsoom Shaikh, Khalid Saifullah Rajput Ghulam Sarwar Gachal, Hissamuddin Bhatti, Muhammad Taha Bhutto, Iqra Raees Shaikh and Hira Lakho

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The yellow-footed green pigeon (*Treron phoenicoptera*) is a member of order Columbiformes under family columbidae. It has an extremely limited distribution, and habitat fragmentation is blamed for that. This species is seen very rarely in different parts of Pakistan and therefore very little is known about its morphology and distribution. In this context, a study was proposed to delve into *T. phoenicoptera* in different areas of Sindh including Mir PurKhas, Badin, Hyderabad and Jamshoro districts from January to September, 2021. The extent of its distribution was

confirmed along with ascertaining the status of bird as permanent resident or temporary visitor in particular areas. Present study recorded Yellow footed green pigeon through thorough observations carried out from early morning to late evening. The bird species was examined for morphological characteristics and all the possible variations in morphological parameters were recorded for the definite systematic arrangement of species in question. Present study recorded the distribution of *T.phoenicoptera* in district Badin and MirpurKhas, wherein district Hyderabad and Jamshoro there was no occurrence of this species throughout the study period of nine months. The body of *Treron phoenicopterus* was recorded as yellowish olive-green, while the crown was blue grey. The outer ring of iris was pink-red and inner ring was observed pale blue. Their bill was silver grey with light green color, while legs were bright yellow. The morphometric of species was recorded as: body weight (g) 251.8±9.3, body length (cm): 29.9±2.5, tail length (cm):9.1±2.1, and wingspan length (cm):19.2±2.0. Distribution of *T. phoenicoptera* was recorded rare in Sindh province of Pakistan.

#### QUANTIFICATION OF MICROPLASTICS IN AN AQUATIC FOOD CHAIN

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Today Microplastics (MPs) are one of the potential threats and emerging environmental pollutant. In last few years, significant research has been done on fresh water microplastics. Although various studies have reported microplastic pollution in rivers of different countries, still numbers are marginal as compared to studies done in marine water bodies. In this study MPs quantification was done in two fresh water bodies i.e. River Ravi (Testing environment) and a university pond (Controlled environment) to trace MPs along the food chain including biotic and abiotic components. Samples were taken from air, water, sediments, planktons, fish and avian specimens from both water bodies. Higher MPs were found in all samples taken from river Ravi ranging from  $3.0 \pm 1.58$  MPs items in water to  $15.20 \pm 3.35$  MP items in air as compared to  $2.8 \pm 1.79$  MPs in water to  $11.20 \pm 1.89$  MP items in University pound respectively. The mean value of MP items in the GIT of all species was higher (5.05 + 2.25) as compared to the respiratory tract (1.57 + 1.3) suggesting ingestion as main mode of exposure. However, this mode of exposure needs to be further investigated along with other exposure routes.

### INSETS; THE SAVIOUR OF ECOSYSTEMS AGAINST POLYSTYRENE

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The planet Earth is getting polluted day by day. Among all the pollutants, polystyrene is the most important. Use of polystyrene in appliances, automotives, medical and electronic industry is crucial. Studies have declared that polystyrene or Styrofoam is safe for use in contact with food. Food packaging industry totally relies on it because of its non-conducting property for heat. Due to this, freshness of food lasts for longer periods. Along with all these benefits, Styrofoam has also a non degradable attitude. Even temperature at 80 °C doesn't affect it at all. This behavior of Styrofoam makes it a problem for the environment. So, we are in dire need to solve this problem of degrading it. For that, Mother Nature comes to our help. Scientists have proved that there are many insect species that can degrade this nasty material effectively. Among those species, wax moth (Galleria mellonella), mealworms (Tenebrio molitor), lesser mealworms (Alphitobius diaperinus), confused floor beetle (Tribolium confusum), super worm (Zophobas morio) and many other members of family Tenebrionidae of order Coleoptera. Different studies has

shown that when larvae of wax moth was left with the polystyrene, worm holes started to appeared in that Styrofoam just after 40 minutes. Talking about the mealworms, Styrofoam was effectively degraded by its larvae under 24 hours and when compared to those larvae which were fed with normal diet, both did equally well. Another study showed that 100 larvae of *T. molitor* can eat upto 34-39 mg of Styrofoam a day. This is a very pleasant news for the environmentalists because Styrofoam is polluting the environment especially the beaches and shores. Another member of the Tenebrionidae i.e. *Alphitobius diaperinus* also have ability to degrade the packing and utensil polystyrene. However, there is much more work needed to be done on tenebrione beetles to explore more insects that can degrade this non degradable product. After degrading the Polystyrene, these insects larvae especially mealworms can also be fed to the poultry. So, these insects serves two purposes, degrading the non degradable and as a feed for our food i.e. poultry. Hence, insects saved the day, again.

## **SECTION - VI**

### **POSTER SESSION**

# NEW RECORD OF FAMILY LIBELLULIDAE DRAGONFLIES (ARTHROPODA: ODONATA) FROM DISTRICT JAMSHORO, SINDH

#### Jaweria Shaikh, Naheed Baloch, Barkat Ali Bughio and Sidra-tul-Muntha

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Dragonflies are valued as a predator due to their widespread predatory nature dragonflies are considered significant biological control tool may help control the population of that type of insects which are harmful like mosquitoes. Dragonflies are beneficial for our environment and our health because they eat harmful insects like mosquitoes, flies, bees and other harmful insects. They usually found on river sides and helping to save atmosphere for humans by feeding on harmful insects. Current study was conducted on new record of family Libellulidae dragonflies collected from the different areas of District Jamshoro, Sindh. A total 228 specimens were collected and identified into 3 genera covering 3 species of dragonflies present in District Jamshoro namely *Bradinopyga geminata* (Rambur, 1842) (16.75%), *Orthetrum sabina* (Drury, 1770) (19.55%), *Chrocothemis servilia* (Drury, 1773) (27.37%). The aim of study is to provide the awareness of dragonflies for the local people from District Jamshoro.

# CONTAMINANT INDUCED DNA DAMAGE IN PERIPHERAL BLOOD ERYTHROCYTES OF OREOCHROMIS NILOTICUS

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Aquatic environment serve as last receptacle of industrial and agricultural effluents which contain highly toxic contaminants in the form of complex mixtures. Among different contaminants, pesticides are highly toxic due to their persistence, higher bio-accumulative potential and lipophilic nature. Therefore, exert adverse effects on non-target organisms. Keeping in view the toxic nature of pesticides, an experiment was planned to assess the DNA damage in peripheral blood erythrocytes of fish, Oreochromis niloticus under chlorpyrifos and bifenthrin pesticides mixture exposure for the period of 30 days. In the first step, 96-hr LC<sub>50</sub> value of pesticide mixture was calculated for O. niloticus in a static system. To check the dose dependent DNA damage, O. niloticus were exposed, separately, to four sub-lethal concentrations of mixture (chlorpyrifos+bifenthrin) viz. 1/3<sup>rd</sup>, 1/4<sup>th</sup>, 1/5<sup>th</sup> and 1/6<sup>th</sup> of LC<sub>50</sub> along with negative and positive control for the period of 30-days. Comet slides were prepared after exposure period by taking blood from caudal vein of fish and examined under Epi-Fluorescence microscope for estimation of DNA damage in terms of %age of damaged nuclei, genetic damage index (GDI) and cumulative tail length of comets (CTL). The %age of DNA damage, GDI and cumulative tail length of comets, induced in peripheral erythrocytes of O. niloticus, due to exposure of chlorpyrifos+bifenthrin mixture varied significantly with the sublethal concentrations of exposure. The various concentrations that induced significant damage to the DNA (%age) and GDI followed the order: positive control > 1/3<sup>rd</sup> LC<sub>50</sub> > 1/4<sup>th</sup> LC<sub>50</sub> > 1/5<sup>th</sup> LC<sub>50</sub> > 1/6<sup>th</sup> LC<sub>50</sub> > negative control. However, incidence of CTL was observed higher due to 1/3<sup>rd</sup> of LC<sub>50</sub> exposure, as evident from their mean value 445.21±0.02 μm while it was significantly lower (3.44±0.05 µm) in negative control. Damage induced by this mixture suggested a serious concern towards their potential danger to the survival of freshwater fish in natural environment of Pakistan. This study also confirmed that the comet assay is a useful technique for assessing the DNA damage in fish.

#### BIODIVERSITY OF ORTHOPTERA FROM DISTRICT SANGHAR

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Orthoptera are cosmopolitan in nature with presence of more than 25 thousands species all over the world. The present study was carried out to investigate the biodiversity of Orthoptera and its important host plants throughout the different field localities such as Hingoro Lake, agricultural fields, and bakeries from different talukas of district Sanghar namely Tando Adam, Shahdadpur, Khipro and Jam Nawaz Ali, during the month of February to July 2018. This study was conducted on the biodiversity of 3 families: Schizodactylidae, Pyragomorphidae, and Gryllidae. Total 375 Specimen were collected with the help of hand picking and pitfall trap method and for the examination and identification were brought in the Advance Entomology Laboratory University of Sindh Jamshoro. During the identification 4 species were discovered under 3 families. The first species *Schizodactylus monstrosus* with lowest (20.26%). Belong to family Schizodactylidae, second *Poekilocerous pictus* with highest of (32.80%) Belong to family Pyragomorphidae, while the two species *Acheta domesticus* with moderate (24%) and *Gryllus integer* (22.93%) Belong to family Gryllidae were found in the district Sanghar. This study will be helpful to begin and guess the vision administration implement for the research in district Sanghar.

# BIODIVERSITY AND TAXONOMY OF BEETLES (COLEOPERA) FROM DISTRICT MIRPURKHAS AND ITS ADJOINING AREAS

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The present study reveals biodiversity (Abundance, richness and evenness) and species composition. Diversity of beetles are wide, they found in all habitat. Beetle contain about 350,000 species worldwide. Beetles are also pest; some are major some are minor pest. Several species are omnivorious, and some are predators. The present study was carried out on biodiversity and taxonomy of beetles (Coleoptera) no proper work has been done in district Mirpurkhas. In order to bring out the knowledge of beetles (Coleoptera) of District Mirpurkhas and seven Taulkas, which is Digri, Hussain Bux Mari, Juddo, Kot Ghulum Muhmmad, Mirpurkhas, Shujabad and Sindhri. There are 400 specimens were collected from various localties of district Mirpurkhas during the year 2017-2018.material were sort out into 7 species belong to 6 genra and 4 families. Family carabidea (2 species), Family Scarabidea (3 species), Family Cerambycidea (1 species), Family Geotruipdea (1 species). These identified species were Anthia sexguttata (4.255%),Carabus hortansis(4.255%), Aneflus calvatus(9.929%), sexguttata Melelontha hippocastine(26%), Melelontha pictrious (21%), Eutheola humilis, (15.95%), Geotrups Stercouious (17%).

# A REVISED CHECK LIST OF MARINE ORNAMENTAL FISHES OF PAKISTAN ALONG WITH NOTES ON COLLECTION TECHNIQUES

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A study of seawater aquarium fish was published in 1991 by authors and now mad some development in it during the last three decades. Therefore, it is necessary to share it with the humanities to take the economics gain

from its trade and obtain welfare by maintaining marine aquarium in Pakistan. In this study 43 species of marine aquarium fishes were reported. In which some of the edible fishes like "Dehndia" etc. was also reported. As some of these fishes are not having abilities of aquarium fishes due to various reasons i.e. highly aggressive, etc. Therefore, in this revised checklist some of these fishes are subtracted. Some new fishes are also collected from Pakistan i.e. neon dottyback fish, Blue tank, etc. The juvenile of some fishes i.e. angle fish are having entirely different colors from the adult coloring. Some of these fishes was double counted in the 1991's checklist. Hence, in this study some of such dispute was also removed. Like Ring Angle have chocolate color body color in the adult age and juveniles are having dark blue body colors. Moreover, in this study notes on collection are also given. As this paper can prove highly economical background for the development of the aquarium fish industry in the country.

# COMMON GYLLIADE ACHETA DOEMTICUS (ORTHOPTERA) AS FOOD SOURCE FOR THE HOUSEHOLD BIRDS

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Acheta domesticus has significant role in dietary. At present we have collected large numbers of Acheta domesticus from district Larkana village Bakrani, and village Bero Chandio from rice field during filed survey we have noticed that many birds particular hen were feed on Acheta doemticus. It might be reason that this species enrich in protein. Present study also recommends that it should be used as protein supplement in future in those countries where is low deficiency of protein.

# FIRST DISCOVERY OF *HIERODULA PATELLIFERA* (AUDINET-SERVILLE, 1839) (MANTODEA: MANTIDAE) CAPTURED A NEW RECORD FROM SINDH, PAKISTAN

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Order Mantodea encompasses the disguised fascinating creatures which are universally recognized as "Mantids" or "Praying mantises" in the insect fauna. They are strictly carnivores, ambush predators with very complex hunting behavior which nourish on immense array of creatures and guzzled them with high speed. Family Mantidae is the prime family that comprises bulk number of ferocious predators which are extensively belongs to the genus *Tenodera*, *Mantis*, *Sphodromantis*, *Hierodula* etc. These entire eyeing 'Alien' like creatures but petrifying pest slayers were captured and recorded as hunters of large number of vertebrates like snakes, lizards, bats, turtles, bats as well as aquatic small guppy fishes. During comprehensive survey for estimating the biodiversity of mantids from district Dadu, for the first time species *Hierodula patellifera* (Audinet-Serville, 1839) has been captured from cultivated fields and were discovered as new record from Sindh Pakistan after China, Taiwan, Java, India, Hawaii etc.

# INVESTIGATION ON SECONDARY DEFENSIVE BEHAVIOR IN STICK INSECTS (ORDER PHASMATODEA)

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Secondary defense mechanism uses after the initiation of prey capture attempt, it include chemical secretion, active escaping, aggression, development of spike, death apparent). When the species feels danger they apply many characters (method) for their defense, Phasmatodea, the so called stick insects defend themselves is their apparent death, In second defense line is their body is made rigid and as the third line they made all their legs parallel to their

body to appear like long and linear stick. In the night time stick insects become active, feeding is also taken where as in the day time they keep rest in food plants and lay with the legs parallel to their bodies, appearing like stick. They coincide their movements, in rhythm with leaves of plants whenever wind blows that is also the part of their defense system. Adult male of stick insects have large spike as secondary defensive structure on their body located at metathoric legs, they are found in group inside hollows in the bases of trees, so they are mostly found in forest areas, stick insects also shown their chemical defense while capturing them the membrane of reproductive organ secrets monoterpene alkaloid actinidine as a defensive chemical, have unwanted odour with brown color thick fluid.

#### SOME INTERESTING FACTS REGARDING CICADAS (CICADIDAE: HEMIPTERA) FROM LARKANA, SINDH

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Cicadas are the most wondering group of insects. They have very unique characteristics. They easily recognized by their loud noise. Its members were captured at 3.30 O, clock of noon which is very preferable time of the species at the villages of district Larkana. The recognized species were collected, identified and preserved in the entomological boxes in Entomological and Bio-Control Research Laboratory (EBCRL), Department of Zoology, University of Sindh, Jamshoro. The measurement and drawing line were taken and their indicative feature was also discoursed. During this study its defense mechanism predator fool hardiness, playing dead, alarm squawk mortality factors were discussed on different host plants along with its management through natural predators.

# COLOR POLYMORPHISM IN PHANEROPTERINAE (TETTIGONIIDAE: ENSIFERA: ORTHOPTERA)

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Color polymorphism is defensive attribute found in Phaneropterinae species which is the most important feature of this group. Phaneropterinae species have one of the most striking behavior of blending of body downward in which the rest of the body remains motionless during the daytime but remain active in night and they perform their most of the activities at the night time because they have less risk of being caught or spotted by other animals such as mammals or birds that are highly dependent on the long horned grasshoppers for feeding purpose. Phaneropterinae species remain susceptible to the predators which cause much danger to them and these species attain the variation by this polymorphic condition to face these predators. Beside this, property of the color polymorphism of individuals are highly heliotherms, in this condition the extreme diurnal condition of body temperature of these species is maintained by the behavioral mechanisms. This research financially supported by Higher Education Commission, Islamabad, Pakistan (Project No. 6737 SINDH/NRPU/R&D).

#### OCCURRENCE OF ACRIDIDAE (ACRIDOIDEA: ORTHOPTERA) FROM THAR DESERT, SINDH

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During the existing survey grasshopper samples were collected from various localities of Thar Desert (Umerkot and Tharparkar). 167 specimens were collected during 2016-2017 from various ecological habitats and sorted out

into single family (Acrididae) pertaining to two subfamilies (Catantopinae and Calliptaminae). Further, specimens were identified into two species *Diabolocatantops innotabilis* and *Acorypha glaucopsis*. In addition to this, taxonomic key along with the complete taxonomic description for each species was also constructed for easy recognition of taxa. Fair collection from this region may lead to the discovery of some new taxon in future.

# BIOLOGY OF *OEDALEUS SENEGALENSIS* (KRAUSS, 1877) FROM DISTRICT HYDERABAD, SINDH, PAKISTAN

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The Grasshopper *Oedaleus senegalensis* is known as pest of economic importance, which cause damage to crops besides pastures of district Hyderabad, Sindh. This study was done to investigate the biology and effect of host plant development on insect under laboratory conditions. Observations in field were made ones/fortnight during March-November, 2018. Total collection was 41 instars and 13 adults from different regions. Collected instars and adults brought to laboratory. Instars reared on rice and wheat leaves in order to observe developmental stages and development time period. On rice mean of instars development was 28 days. 1<sup>st</sup> to 5<sup>th</sup> instar development time period 4,6,5,7 and 6 days respectively, while on wheat it was 4, 4,6,5,5 and 6 days respectively for 1<sup>st</sup> to 6<sup>th</sup> instars and total life spawn was noted 30 days. Scanning Electron Microscopy (EDS) of rice leaves was also done.

#### OUTLOOK OF PHANEROPTERINAE (TETTIGONIIDEA: ENSIFERA: ORTHOPTERA) OF SINDH

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Phaneropterinae species do their maximum activity at the night time specially the reproductive time of most of the species starts at night after spending a long daytime at thick vegetation on trees and shrub leaves. During the present study 232 specimens were collected and sorted out into 04 species i.e. *phaneroptera falcate* Poda, *Phaneroptera quadripunctata* Brunner von Wattenwyl, *Pseudonareta robusta* Bei-Bienko, *Anisotima dispar* Bei-Bienko. It was noticed that Phaneropterinae inhabited in various ecological zones and cause huge damage to agricultural lands. Its oviposition and copulation behavior is very interesting and attract much attention of entomologist at the present its various biological aspect are under our study will be presented in future.

# REGIONAL RECORD OF FIELD CRICKETS (GRYLLIDAE: ORTHOPTERA) FROM DISTRICT MIRPURKHAS, SINDH

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Mirpur Khas district lies in lower region of Sindh province, which is located in the south-east part of Pakistan. New regional record of species *Gryllus multipulsator* is recorded from district Mirpur Khas. This species was first reported by Weismenn in 2009. It is mainly found in South California, South Nevada, South west Arizona and the pacific slopes of Mexico. During present study, 8 specimens consisting 5 males and 3 females were found. Specimens were collected from Digri, Mirpur Khas. We have examined that *Gryllus multipulsator* is found very closely related to *Gryllus assimilis* however it differ due to following characteristics: pubescent pronotum, head narrower than pronotum, hairs at the junction of head and pronotum and hind tibia with 5 spines. Present study

recommends that if more extensive survey would be carried out there is expectancy of more new records and species from this region.

# STUDY ON COLOR DIMORPHISM AND CAMOUFLAGE IN ACRIDINAE (ACRIDIDAE: ORTHOPTERA) FROM DISTRICT MATIARI

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Present study was carried out to capture the short - horned grasshoppers from different localities and agricultural fields of district Matiari. As a result of this study, a total of 350 specimens were collected and identified from cultivated and uncultivated areas. Sorted out four species i-e *Acrida exaltata* (Walker 1859), *Truxalis fitzgeraldi* (Dirsh 1950), *Phlaeoba tenebrosa* (Walker 1871), *Duroniella laticornis* (Krauss 1909) and a single sub-species *Truxalis eximia eximia* (Echwald 1830). These graminicolous hoppers have color dimorphism mechanism and camouflage pattern, sometimes very difficult to captured Acridinae hoppers due to present of these both factors. They have ability to rapid change their body coloration and blend with surrounding. These both factors adapted for against the predation and easily escape their enemies. Acridinae specimens appear in two different forms as size and color (dimorph) in species of same population, in some species male specimens have bright colored wings for attraction of female specimens.

# DESCRIPTION OF NEW TREMATODE *PARATANAISIA HAMALI* N.SP. (TREMATODE: EUCOTYLIDAE) IN MALLARD *ANASPLATYRHYNCHOS* (ANSERIFORMES: ANATIDAE) OF SINDH, PAKISTAN

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In the result of ongoing helminthological studies of Mallard Anasplatyrhynchos of Kambar Shahdadkot District of Sindh Province, Pakistan, and a total of 18 birds were captured from different localities. During the examination of gut contents and visceral organs, 27 specimens of Paratanasia hamali n.sp. collected from the kidney of the host bird. Paratanaisia hamali n.sp. differs from its close allies in body shape and size, distribution of vitellaria commencing from intestinal fork situated in lateral fields of the body, the presence of seminal receptacle, shape of testes, ovary and eggs scattered in whole body. On the basis of these diagnostic differences, a new species Paratanaisia hamali is proposed. Previously this genus was reported by Buriro et al., 2016 from bird Anasplatyrhynchos of Pakistan. However, this is the second report of the genus Paratanaisia from Pakistan.

# INCIDENCE OF INTESTINAL HELMINTH PARASITES (CESTODA: CYCLOPHYLIDAE) ON VARIOUS BREEDS OF CHICKEN *GALLUS GALLUS* (AVES: GALLIFORMES) FROM DISTRICT HYDERABAD, SINDH, PAKISTAN

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Poultry are domesticated birds which are kept by the humans for their eggs, meat and for their feathers. The chickens *Gallus gallus* (Linnaeus, 1758) are one of them, is medium size birds characterized by fleshy red combs and wattles on their heads. Males are usually larger and called as cocks, and have more inflated plumage than

females which are called hens. The chickens are omnivorous and ground-dwelling birds. Their feeding habits make them infected from various types of ecto and endoparasites like Protozoan Arthropods, and helminthes. The chickens *Gallus gallus* (Linnaeus, 1758) were purchased from the market of Hyderabad city regularly and brought to the Advanced Parasitological laboratory for processing. The study was conducted from January 2019 to October 2019. The chickens were dissected and their intestine were carefully removed and examined for helminth parasites. The cestodes parasites were collected from the intestine preserved in viles contained 70% Alcohol, stained with borax carmine and finally mounted in Canada balsam. The parasites were examined and identified under the microscope. Out of 15 chickens, 13 were found infected with helminth parasites. Presently 52 cestodes parasites were recovered belonging to 03 genera and 3 species. There is no trematodes and nemateodes were recovered. The species and their incidence are 46.15% for *Choanotaenia infundibulum* (Bloch, 1779), 30.76% for *Raillietina cesticillus* (Molin, 1858) and 23.07% for *Cotugnia digonopora* (Pasquale, 1890). Consequences of this study indicate that the damage caused by these parasites bring about pathological and metabolic changes in the intestine which definitely affect the physiological functions of the organs and the vital activities of the chickens. This was concluded through examined results that extreme damage caused by these parasites brings about specific socio economic losses in all aspects.

#### STUDIES ON VARIETY OF BIRDS (CLASS AVES) INHABITING SINDH PAKISTAN

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Birds are the flying vertebrates that are over 10,000 species distributed worldwide. Each avian species possesses diverse form and plays vital role in the ecology; therefore, present study was proposed to explore Sindh province for recording the morphology, nesting behavior and ecological significance of diverse species of birds including *Passer domesticus*(House sparrows), *Sterptopeliadecoacto* (Eurasian collared dove), *Amourornisphoenicurus* (white breasted waterhen) and *Columba livia* (Rock dove). For the collection of data, different areas including District Naushahroferoze, Mirpurkhas and Hyderabad were surveyed randomly from July to November, 2019. The study sites were marked for sampling and observation, whereas electric weight machine, metric ruler, divider, Vernier caliper etc. were used for morphometric of avian specimens. The related scientific literature was also reviewed to determine past and present status of avifauna and interviews from local people were taken for the identification of bird species. The variation in morphological characteristics of male and female of each species was examined along with observation of nesting behavior demonstrated by both sexes (male and female). Parental care exhibited by both sexes was also recorded during each survey. Present study recorded the important role of each species: *P. domesticus*, *S. decoacto*, *A. phoenicurus* and *C. livia* necology. It was thoroughly recorded that the bird species play positive role more than their negative aspects of the ecosystem and even they are very important for keeping ecosystem within suitable limits.

# STUDIES ON WHITE BREASTED WATERHEN INHABITING DISTRICT NAUSHAHRO FEROZE, SINDH, PAKISTAN

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Amaurornisphoneicurus (White breasted waterhen)belongs to Rallidae family of Order Gruiformes. This species is found near wetlands especially of brackish water. The main threat to the survival of this specie is destruction, modification and fragmentation of the natural habitat. Therefore, they are protected in different regions of world as in Maldives. For present study, this species was collected near water bodies in agricultural are as district Naushahro Feroze. Different habitats including wetland, grassland, cultivated land, and human habituations were found embracing the A.phoneicurus. The morphology of A.phoneicuruswas examined as followed: body weight 285-300 gm, body length 28–32 cm, bill size 3.5-3.8 cm, length of wings 12-13 cm, length of tail wing 5-6 cm. Tarsus were measured 6 -7 cm, while middle toe was about cm. White breasted waterhen is used as source of food and

therefore it is overexploited in various regions of the world. Therefore, there is need of conserving the population of *A.phoneicurus*in wild as it plays major role in maintenance of aquatic ecosystem and also keeps population of invertebrates within limit as it is active predator of invertebrates.

### STATUS OF *CHELONIA MYDAS* (GREEN SEA TURTLE) AND HABITAT AT HAWKE'S BAY, KARACHI, SINDH, PAKISTAN

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Chelonia mydas (green sea turtle) is among the five turtle species that exist in Pakistan and it is listed as endangered species in IUCN red list. Considering their endangered status, present study was proposed to record current status of Chelonia mydas as well as its main habitat "Hawke's bay" at Karachi. Several surveys were carried out in between sunset to late night from August to November for observing the nesting condition of green sea turtles and rate of pollution along their habitat. The nesting behaviour was recorded during night only, however status of nests and habitat was observed during both day and night. Information about habitat of Chelonia mydas (green sea turtles) was also collected from the Turtle conservation unit. The results showed that green sea turtle faces wide variety of problems such as high rate of pollution, existence of large number of predators, human encroachment and disturbance at Hawke's bay Karachi. Feeding on plastic materials was observed as one of the main causes of their deaths. Sea turtle were observed to get choked to death due to consuming plastic bags and water bottle caps. Numerous nests of green sea turtles were found deteriorated by the predators like dogs, crows, skunks etc. Due to lack of conservation efforts, the green sea turtles were observed facing the loss of habitat and climate change as well. Green sea turtle was observed being affected by the large number of light plastic and other marine debris at Hawke's' bay. Besides that, sound pollution, thermal pollution and chemical pollution also threat to marine life as well the terrestrial life of turtle at the beach. Therefore, there is need of increasing the awareness among local communities about the importance of green sea turtle as they are easily susceptible to anthropogenic activities at every stage of their life. In this context, it is very important to implement strict conservation rules for the conservation of nests and the juveniles of green sea turtles. Implementing the conservation efforts and educating the local people are the serious urge for the conservation of green sea turtles at Hawke's bay.

#### ANTIMICROBIAL ACTIVITY OF GUT MICROBIOTA OF HONEY BEE (APIS CERANA)

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The aim of present study was to investigate the antimicrobial activity of Honey bee gut microbiota. Honey bees were collected from local bee keeper from District Lahore, Punjab, Pakistan. Bees were dissected and total 12 strains were isolated and purified from HB gut homogenate. These strains were then characterized morphologically and biochemically. Non motile Gram positive strains were screened for their antimicrobial activity by primary screening against *E. coli, P. aeruginosa, K. pneumonia, B. licheniformis* and *Salmonella* Spp. using cross streak method. In preliminary screening HB gut isolate B1 showed highest zones of inhibition (ZI) against *E. coli* and *P. aeruginosa* (11mm and 10mm ZI) respectively. Isolate B5 showed highest activity against *K. pneumonia, B. licheniformis* (8mm ZI). M4 showed highest activity against *P. aeruginosa* which was (7mm ZI) respectively. B1, B5, B6, M4, M5 and M7 showed good antimicrobial activity and were further subjected for their secondary screening using agar well diffusion method. Cell free supernatant was obtained by culturing bacterial isolates for 48 hours, centrifuged and filter sterilized. Antimicrobial activity using agar well methods revealed that among all isolates, M4 showed highest inhibition against all pathogens. Only isolates M4 and M7 inhibited the growth of *P. aeruginosa* with ZI of 11mm and 8mm. Isolate B1 showed highest inhibition against *E. coli*, with 13mm ZI. After secondary

screening, antibiotic susceptibility testing of two isolates (B1 and M4) with promising antibacterial activity was done and they were subjected to physiological and genetical characterization. 16S rRNA sequencing revealed these isolates as *Bacillus tequilensis* and *B. cereus*. Our findings suggested that HB gut microbiota have broad range antimicrobial spectrum and this could be used as an alternative to antibiotics.

### A COMPARATIVE STUDY OF DIVERSITY AND EVENNESS INDEX OF AVIAN FAUNA OF UCHHALI WETLAND COMPLEX AND KALAR KAHAR LAKE

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A study was conducted to study the diversity and evenness of avian fauna of Uchhali Wetland Complex and Kalar Kahar. During the survey total 53 species are observed and recorded. Diversity index and evenness index of each lake was calculated. Jahlar has the highest diversity index of (2.69) because of smallest area of 17 hectares while Kalar Kahar has the second highest diversity index of (2.23). Jahlar Lake has the highest evenness index of (0.83) while Kalar Kahar Lake has second highest evenness index of (0.6).

# ASSESSMENT OF HUMAN-HERPETOFAUNA INTERACTION AND THE DIVERSITY OF HERPETOFAUNA IN DAPHAR FOREST SANTUARY, DISTRICT MANDI BAHAUDDIN, PUNJAB-PAKISTAN

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This survey based study was conducted in different areas of District Mandi Bahauddin including Daphar forest Sanctuary, rural areas, urban area and agricultural area. The main purpose of study was to find diversity of herpetofauna in these areas. Daphar forest has 7812 acres area. Total 28 species belonging to five orders and 14 families were recorded and used. Out of total 28 species, 26 were recorded from forest, 19 were recorded from rural, from agricultural area 18 species were recorded and 2 species were recorded in urban area. Highest number of individuals is recorded from forest 126 and onward population decreased as; rural (94), agricultural (46) and urban (27). Simpson diversity (0.94) and Shannon diversity (3.01) in forest habitat was high as compared to other habitats. Margalef Indices also show that (5.17) value from forest depicts availability of high diversity than other areas. Frequency of citation (FC) obtained during survey in district Mandi Bahauddin shows that *Hemidactylus flaviviridis* has highest diversity with highest FC value of 41 and *Acanthodactylus cantoris* has lowest FC value 01.

#### ASSESSMENTS OF AVIAN DIVERSITY OF UCHHALI LAKE, PAKISTAN

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The wetlands of Pakistan covers a variety of ecosystems. Wetlands are the main source to country economy of Pakistan as they provide a site for tourism and important habitat of many valuable species of birds. The objective of the study was to know the avian diversity especially of aquatic avian fauna at Uchhali Lake from October 2015 to

September 2016 on monthly basis. During the research total 36 avian species were observed in the vicinity of Uchhali Lake with a population of 13,342. Out of total, common coot was the top populated species and population was noted as 8510. Among the 36 species observed at the Uchhali Lake 56% were noted as resident, 39% were noted as winter visitor, 5% were noted as summer breeders. During the research total 36 avian species were recorded in the vicinity of Uchhali Lake, and population was 13,342. Common coot was the most abundant species, out of total avian species, population was recorded as 8510 and relative abundance was 63.78. While common pochard was 2nd dominant species with the total count was 1275 and relative abundance (RA) was 63.78. Avian species with the lowest number was recorded as crested lark.

### ASSESSMENT OF MAMMALIAN DIVERSITY IN THE TERRITORY OF DAPHAR FOREST, DISTRICT MANDI BAHAU DIN, PUNJAB PAKISTAN

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The present research was conducted to evaluate the mammalian diversity of the study area, and to explore the level of human-mammals interaction along Daphar forest, Pakistan. The mammalian diversity was recorded along forested scenes, agricultural, semi-urban and urban territories. Throughout the year a total of 20 mammalian species were recorded from nearby areas of Daphar forest. Linear count method was applied to estimate the populations of various mammalian species and diversity was measured through Shannon-Wiener index. A Total 20 species were recorded, out of total 18 were recorded from the forest, Shannon-Wiener diversity index for SA1, SA2, SA3 and SA4 was 2.653, 1.627, 2.188 and 2.132 respectively while the dominance was recorded as 0.0788, 0.207, 0.1247 and 0.1422. High Shannon-wiener Index (2.653) in forest and high richness (3,144) shows high diversity in forest as compared to other habitats.

### COMPARISON OF MORPHOMETRIC PARAMETERS IN WILD AND DOMESTIC PIGEON FED ON PEARL MILLET (PENNISETUM GLAUCUM) DIET

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The experiment was conducted to characterize domestic and wild pigeons. Twenty five domestic and twenty five wild pigeons were reared in controlled conditions. The following morphometric parameters such as mean body length, body weight, wingspan, shank length, bill length, tarsus length and head length were measured from week 1 to week 26 in wild and domestic pigeons. The mean body weight of domestic pigeon from week 1 to week 26 was recorded as  $(88.10\pm2.81)$  to  $(287.50\pm0.39)$  while in wild pigeon was recorded as  $(92.00\pm2.83)$  to  $(288.5\pm0.24)$ . Wingspan of domestic pigeon from week 1-26 was recorded as  $(8.01\pm0.21)$  to  $(68.68\pm0.03)$  while in wild pigeon was recorded as  $(8.08\pm0.21)$  to  $(68.68\pm0.03)$ . Bill length of domestic pigeon was recorded as  $(0.79\pm0.02)$  to  $(1.84\pm0.00)$ . Hence bill length of wild pigeon was recorded as  $(0.86\pm0.02)$  to  $(1.82\pm0.00)$ . Our study depict that there is significant difference exist in morphometric parameters of domestic and wild pigeons fed on pearl millet diet.

# A NEW RECORD AND NEW SPECIES OF THE GENUS *ACUARIA* BREMSER, 1811 (ACUARIIDAE: NEMATODA) FROM SINDH JUNGLE SAPRROW (PASSERIDAE: PASSERIFORMOES) IN LARKANA, SINDH, PAKISTAN

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During the present research investigation for the helminth parasites, total fourteen *Passer pyrrhonotus* (Sindh jungle sparrow) were examined from Larkana districts of Sindh, Pakistan. Out of fourteen birds only one were found infected with nematodes. Five nematodes of genus *Acuaria* Bremser, 1811 (023,03) belonging to Acuariidae: Nematoda were collected from intestine of *Passer pyrrhonotus*, killed and preserved in 70% Ethanol and Glycerol solution for detailed study. Diagrams were made with the help of camera lucida and photographs were taken with Nikon digital camera. Measurements are taken in millimeters (mm) and of eggs in micrometers ( $\mu$ m). The specimens were compared with the different species of genus *Acuaria*, reported from different birds of around the world. The new species is named as *Acuaria pakistanensis* n.sp. The name of new species refers to the country name from where it was collected. However, this genus and host *Passer pyrrhonotus* (Sindh jungle sparrow) is first time reported from Pakistan.

### SPECIES DISTRIBUTION AND RED DATA LIST OF JUMPING SPIDERS FROM PAKISTAN

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Spiders have invaded every habitat on the Earth except polar region, highest mountains and oceans. They play an important role in ecosystem as biological indicator for the changes in habitat quality. Along with that their role in pharmaceutical industries and agroecosystem are under investigation. Among spiders, Family Salticidae is rich in diversity and consists of 6115 species that belong to 636 genera. However, their inventory from Pakistan is incomplete and required attention. Till now only thirty three species of salticid have been reported from Pakistan. Of these, status of nineteen species is dubious. Biogeography and distribution ranges of salticids have never been reported from Pakistan. For this study, a total of 2732 specimens collected from different areas of Pakistan using visual search and netting methods. During surveys, GPS points (longitude, latitude and altitude) were noted for each collected specimen. Bioclimatic profiles were used to assess their potential distribution. BioClim and Maxent model in the Arc GIS were used to predict the potential distribution of the recorded genera of salticids in Pakistan. On the basis of their actual and predicted distribution ranges, 25 recorded species were widespread, four moderately distributed and three restrictively distributed. Among the studied species four were new to science, 23 recorded first time from Pakistan. Species endangered according to IUCN is also recorded from two areas of Pakistan

# NEW RECORD OF GENUS *BRACHYLECITHUM* SHTROM, 1940 IN *VANELLUS LEUCURUS* (CHARADRIIFORMES: CHARADRIIDAE) OF HYDERABAD, SINDH, PAKISTAN

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During present study on abundance of helminth parasites of *Vanellus leucurus*, twenty specimens of trematodes of genus *Brachylecithum* were collected from gall bladder of the host. Body of the specimens is elongated, cylindershaped, forebody slightly narrow than hind body, which is rounded anteriorly tapering posteriorly; oral sucker circular; ventral sucker round protruded outside of the body; prepharynx absent; Pharynx spherical, broader than

long; vitellaria post-ovarian in position, commencing near ovary, limited in extent; esophagus cylindrical, tubular, medium; testes larger than ovary, oval, tandem in position; ovary small, round, slightly wider than long, sub median, post-testicular; uterus commence from the level of ventral sucker, occupying all the hind body, posterior to vitellaria; eggs numerous, dark brown in color. On the basis of these diagnostic characteristics present specimen has close resemblance with *B. americanum* in all the crucial structures and identified as such. Various species of genus *Brachylecithum* are reported from different parts of the world from variety of hosts. However, this genus described first time from the host *Vanellus leucurus* in Pakistan, making it a new host record.

### ANALYSIS OF FRESHWATER FISH FOR INTESTINAL HELMINTH PARASITES, IN RIVERS OF DISTRICT CHARSADDA, PAKISTAN

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The present study was conducted on the analysis of intestinal helminth parasites of selected freshwater fish collected from the three rivers (Sardaryab, Khyale, Jendi) of district Charsadda. A total of 300 samples (100 from each river) were collected in summer (June, July and August). These samples were then analyzed for morphometric measurement of fish, identification of heminth parasites and determination of the population dynamics of identified helimth parasites. Overall out of these 300 samples, 175 were found infected. A total of 15 genera of helminth parasites have been identified. Among these genera 05 were cestodes, 05 trematode, 02 nematode and 03 acanthocephalans. It is evident from the current investigation that among the recovered helminth parasites cestode has a maximum prevalence (52%), intensity (39) and density (0.44) in all the fish samples of the three rivers followed by trematodes (prevalence 25%, intensity 27 and density 0.31) nematodes (prevalence 14%, intensity 13 and density 0.15) and acanthocephalan (prevalence 09%, intensity 06 and density 0.06). For all the intestinal helminth parasites, the highest prevalence, was observed for river Sardaryab (65%) followed by Khyale (58%) and Jendi (51%). The highest intensity among the four fish species of the three rivers was observed for Cyprinus carpio (2.93) followed by Ctenopharygedon idella (2.23), Tor putitora (1.52) and Clupisoma naziri (1.16). A maximum (0.94) density is obtained for Cyprinus caropio and minimum (0.94) for Clupisoma naziri, whereas 0.82 for Tor putitora and 0.78 for Ctenopharygedon idella. It is evident from the obtained results that there is a direct or indirect impact of different environmental factors and feeding habits on the population dynamics of intestinal helminth parasites. For all helminth parasites, high intensity and density has observed in June and July while lower for the samples collected in the mid and late August which is the monsoon season in Pakistan. This indicates that elevated temperature, adequate moisture and less rainfall are essential for parasites growth.

## PARTITIONING OF SOME SELECTED HEAVY METALS IN ENVIRONMENT OF AJK, PAKISTAN

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Current study focused on some selected heavy metals in ground water, stream water, soil, human nails and hair samples collected from selected sites of the state of Azad Jammu and Kashmir (AJK). Special dispersal patterns of heavy metals elements exposed not only the possible impacts of density of human population and topographies of the soil and associated events contribute to ecological pollution through the heavy metals in AJK. The concentration of heavy metals detected in all the soil samples found below than standard guidelines set by European Union (EU) for soil. In the samples of ground water, stream water, higher concentration of heavy metals recorded than the standards guidelines suggested by World Health Organization (WHO). In human nails and hair samples the concentration of

heavy metals also calculated above the safe guidelines advised by, England, Italy and Japan. The water and human being are receiving metal contamination due to several factors in the study area most remarkable are the human activities for agriculture, domestic/industrial discharge of waste water into the river without any treatment and road developments projects. Heavy metals through river water can make a big risk to human health and aquatic life by the process of bioaccumulation and bio-magnification.

### FUSARIUM EQUISETI ISOLATED AND CHARACTERIZED FROM SILVER CARP, HYPOPHTHALMICHTHYS MOLITRIX

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Fusarium species were stated to be the natural pathogens of freshwater fishes. Fusarium infection was identified from silver carp reared in earthen ponds in Punjab. Microscopic studies were conducted on material taken from infected silver carp. Hyphae, colony pigmentation and chlamydospore formation and the presence of macrospores and microspores confirmed that the isolated fungus as Fusarium equiseti. Isolated Fusarium was confirmed by internal transcribed spacer (ITS) sequencing which matched (100%) to other F. equiseti sequences available in GenBank. To check the pathogenecity, experimental injection of F. equiseti were administered into silver carp fingerling. The result showed development of fungal mycelium on the fish skin and gills. This challenge infection showed that F. equiseti is pathogenic to silver carp and fish showed 80% mortality. Histopathological observations of stained sections of various organs of moribund fishes revealed pathological changes of different severity in organs including gills, liver, heart, kidney, intestine and muscle. Pathological changes observed were: atrophy, hypertrophy, hyperplasia, necrosis, inflammation, steatosis, fibrosis, metaplasia. Gills were the most affected organ of the fish. Fungal hyphae were also observed in the muscle of post-injected fish. This is the first report of F. equiseti from silver carp reared in earthern ponds. It appears as an emerging pathogen, thus may pose significant risk on health of freshwater fishes in the world.

# HISTOPATHOLOGY OF *TAENIA PISIFORMES* BLOCH 1780 (CYCLOPHYLLIDAE: TAENIIDAE) EXTRINSIC PARASITE OF LIVER IN INFECTED RAT (*RATTUS RATTUS*) FROM LARKANA, SINDH, PAKISTAN

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A study on the Histopathology of *Taenia pisiformes* (Cyclophyllidae: Taeniidae) was carried out at the advance laboratory of Parasitology of the Department of Zoology, University of Sindh Jamshoro during the year 2016-2017. A total of 10 brown Rats *Rattus rattus* were examined for their helminthes parasites. Only 02 Rats out of 10 were infected with *Taenia pisiformes* on liver in the form of bladder. Tissue samples from infected parts of the liver were fixed in Bouin's fluid. The series of sections were made by microtome technique and were mounted in Canada balsam. In the histopathological findings, infected liver of Rats revealed mononuclear and polynuclear cellular infiltration in the necrotic area, fatty degeneration and areas of coagulation necrosis of the hepatic cells. The liver had congested blood vessels and congested sinusoids.

## ABUNDANCE OF PHYTOPHAGOUS SCARAB BEETLES (COLEOPTERA: SCARABAEIDAE: MELOLONTHINAE) IN HYDERABAD AND ADJOINING AREAS, SINDH

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Phytophagous scarab beetles belong to order coleoptera, family scarabaeidae and sub-family melolonthinae. They are exclusively phytophagous and also named chafers. Adults may attain the size upto 3-58 mm with black or even reddish brown coloration. Head devoid of horns equipped with well developed mandibles .Adults as well as larvae are highly important because of their destruction to variety of crops, pastures and grassy lands. Majority of them are nocturnal and crepuscular. Fortnightly observations were made and the specimens were caught by multiple methods (mercury light trap, pitfall trap and hand picking) for six months starting May 2018 to October 2018. Total 1455 specimens were collected and identified into three species of genus *Melonatha*.

### THAPAROTREMA AKBARI NEW SPECIES OF GENUS THAPAROTREMA GUPTA, 1955 FROM HOST RITA RITA (SILURIFORMES: BAGRIDAE) OF RIVER INDUS, SINDH, PAKISTAN

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During current study on group of ray-finned fishes belong to order Siluriformes, *Rita rita* catfishes collected from River Indus Jamshoro, Pakistan. These host were dissected for examined of helminth parasites. A total of 12 specimens of genus *Thaparotrema* were collected from gallbladder of 9 infected hosts. The new species *Thaparotrema akbari* identified on the basis following differential characters having large body size, elongate and narrow shape, oral sucker elongate and egg shape, ventral sucker wheel shaped, pharynx oval shape, anterior testis rounded to quadrangular in shape, posterior testis pentamerous in shape with broad anterior and narrow posterior end, ovary heart shape, Seminal receptacle tube shape and vertical in position, uterine coils almost overlapped caecum and whole middle region of body. The name of new species refers to the honored of author's father name Soofi Akbar.

# FEASIBILITY ASSESSMENT FOR THE SALMO TRUTTA FARIO (BROWN TROUT) AND ONCORHYNCHUS MYKISS (RAINBOW TROUT) IN RIVER LUSPUR AND LOTKHO, DISTRICT CHITRAL KHYBER PAKHTUNKHWA PAKISTAN

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The current study was carried out to investigate the physicochemical parameters and assess the feasibility for the *Salmo trutta* fario (Brown trout) and *Oncorhynchus mykiss* (Rainbow trout) in River Luspur and Lotkho, District Chitral KP Pakistan. For this purpose, the physicochemical parameters such as pH, electric conductance, temperature, total dissolved solid (TDS), total hardness, alkalinity and chlorine contents of water samples collected from River Luspur and Lotkho, located in District Chitral using standard procedures. The investigated values were compared with the standard values found earlier for optimum growth and productivity of *Salmo trutta* fario (Brown trout) and *Oncorhynchus mykiss* (Rainbow trout). In results, it was found that the average values of River Luspur were pH = 7.88, electric conductance = 0.447 mS cm<sup>-1</sup>, temperature = 11 °C, TDS = 63 mg/L, hardness = 120 mg/L, alkalinity = 74.3 mg/L and Cl-1 = 11.16 mg/L were recorded. Similarly, in case of river Lotkho, the average values regarding

different physicochemical parameters were: pH = 7.71, electric conductance = 0.448 mS cm<sup>-1</sup>, temperature = 8.5 °C, TDS = 61.5 mg/L, hardness = 114 mg/L, alkalinity = 104.3 mg/L and Cl-1 = 9.83 mg/L. In conclusion, the physicochemical parameter of both the rivers were slightly different from each other. Moreover, these parameters were found within the normal range for optimum growth and productivity of the studied trout species.

# NEW RECORD ON THE SPECIES OF GENUS *CONDERUS* ESCHSCHOLTZ (1829) (COLEOPTERA: ELATERIDAE: AGRYPNINAE) FROM KHAIRPUR SINDH, PAKISTAN

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For the current studies the click beetles were collected from the various districts of Sindh, more studies was carried on the Systematics and biogeography of click beetles (Elateridae: Coleoptera) at Entomology laboratory of zoology Department of Shah Abdul Latif University Khairpur, specimens of beetles were collected from the light and various crops field, the specimens identified into single genus *Conderus* (Eschscholtz, 1829)of single species *Conderus mithiensis* (Platia, 2016), the finding of this species constructed a new record for the present area with description, along with genitalial description also provided, Hopefully present study will be for basic tool for the identification of this species.

## PREVALENCE OF GASTRO INTESTINAL NEMATODES IN BUFFALO (BUBALUS BUBALIS) OF TEHSIL KHWAZA KHELA, DISTRICT SWAT, KHYBER PAKHTUNKHWA, PAKISTAN

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The study conducted in 2019 for prevalence of intestinal Nematodes of buffaloes in Tehsil Khwaza Khela, District Swat. A total of 300 faecal samples randomly collected from different sites were examined under microscope for identification of Nematodes eggs. The two types of Nematodes *Oesphagostomum radiatum* 71/300 (23.67%) and *Trichostrogylus spp.* 111/300 (37%) were found prevalent in examined samples. The age wise prevalence of *Oesphagostomum radiatum* was 9/55 (16.36%) in buffalo calfs aged below 2 years and 62/245 (25.31%) in adult buffaloes ages above 2 years, while that of *Trichostrogylus spp.* were 13/55 (23.63%) and 98/245 (40%) respectively. On other hand in bulls the prevalence was 6/25 (24%) of *Oesphagostomum radiatum* and 11/25 (44%) of *Trichostrogylus spp.* In female buffaloes the prevalence of *Oesphagostomum radiatum* was 65/275 (23.63%) and *Trichostrogylus spp.* was 100/275 (36.36%). The Chi-square test analysis of SPSS was applied to find out the prevalence of nematodes on the basis of sex and age of buffaloes.

### FORAMINIFERA AS ECOLOGICAL INDICATORS FOR MARINE POLLUTION IN SANDSPIT

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Foraminifera are found in all marine environments, they are planktic or benthic in mode of life. Foraminifera are calcareous test bearing group in amoeboid protists have shown potential for pollution biomonitoring. Biological indicators or bioindicators provide indication of environmental conditions including presence of pollutants in ecosystem. Biological monitoring enables the finding of impacts and is more connected to ecological health of an

ecosystem than chemical analysis. In the current study we have verified the significance of foraminifera in detecting marine pollution. The foraminifera can resist heavy metal pollution and accumulated organic matter. From the polluted sites presence of stress tolerant taxa, sensitive taxa of foraminifera have been reported. In the current study species of foraminifera, *Ammonia tepida, Cornuspira planorbis, Elphidium sp., Spiroloculina, Quinqueloculina lamarckiana, Cornuspira involvens, Bolivina vadescens etc.*, in Sandspit area are reported. In North East monsoon highest number of Foraminifera was observed in Station II i.e., 11373 individuals/m³ than in South West monsoon Station I, 4458 individuals/m³ and Autumn Inter Monsoon, Station I, 2441 individuals/m³. The peak abundance is in South West Monsoon than in Spring inter Monsoon, Autumn Inter Monsoon and North East Monsoon at coastal waters of Gadani. Abundance of zooplankton varied from 176-12631 individuals/m³.

#### ASSESSMENT OF CALCIUM AND PHOSPHOURS IN YOUNG SUBJECTS

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Several studies showed an inverse association between dietary calcium and phosphorus intake with weight management. The aim of this study is to investigate the association between dietary calcium, phosphorus intake and body mass indices. This cross-sectional study was conducted to investigate the association between dietary phosphorus, calcium intake and anthropometric correlates. 1000 people were randomly selected from different educational institutes of Lahore, Pakistan. Anthropometric measures were calculated, while demographic and dietary information (especially high-calcium and phosphorus sources) was assessed with the help of questionnaire using standard calcium and phosphorus charts. (USDA National Nutrient Database for Standard Reference Release 28). Out of 1000 individuals about 5% were obese, while 33.3% overweight, 53.3% normal and 8.4% were underweight for studied population. Average calcium intake for studied population was 706.66±4.2 mg/d and average phosphorus intake calculated was 747±4.62. The average BMI of 22.84 kg/m<sup>2</sup> was observed in studied population percentage of calcium deficient (<400 mg/day) obese and overweight subjects was found to be 13%. The participants consuming medium amount of phosphorus (i.e. >400) were (29%) males and (36%) were females. High amount of phosphorus (i.e. < 400) was consumed by females (64%) while intake of high phosphorus in males was (56%) that is lower than females. A statistically significant inverse association was observed between dietary calcium intake and obesity suggesting that an increase in calcium intake can decrease the chances of obesity incidence also non-significant inverse association was found between phosphorus intake and obesity. The present study concluded that low intake of calcium and phosphorus may be increases the incidence of obesity.

# BIOLOGY AND MORPHOLOGY OF DIFFERENT STAGES OF SUGARCANE WHITEFLY (ALEUROLOBUS BARODENSIS MASK) IN SINDH

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The development stages of sugarcane whitefly *Aleurolobus barodensis* were studied on sugarcane potted plants in field condition at PARC-National Sugar and Tropical Horticulture Research Institute, Thatta, Sindh. It consists of six stages of life; egg, four nymphal instars and adult. The colour of egg is light yellow conical with a rounded base with a stalk at one end; the eggs changing to dark before hatching, the mean incubation period of eggs were 7.33±1.03, the developmental period of embryo was 06 to 09 days. The newly emerged first instar nymphs were pale yellow in colour, also known as crawler, they slowly change to a shiny black, the first instar lasted for 04-07 days with a mean 5.27±1.19 days. The second instar nymphs were dark in colour and amount of wax on the body with an

oval or elongate-oval body, the mean length of  $2^{nd}$  instar lasted  $5.73\pm1.33$  days and ranged for 04-08 days. The colour of  $3^{rd}$  nymphal instars was also dark in colour with an oval or elongate-oval body and wax cover the body, the  $3^{rd}$  instar duration was 03-08 days with mean  $5.83\pm1.68$  days. The fourth instar is flat, oval, grayish in color with wax over the body and slightly bigger than other nymphal instars and there is 'T' shaped white marking on the thorax. The fourth pupal instar which last 8-13 day with a mean of  $9.23\pm1.43$  days. The adults are dull white, the head abdomen and legs are pale yellow and eyes are red. The adult females lived for 02-06 days with mean  $3.73\pm1.14$  and of the male was 2-4 days with a mean of  $3.00\pm0.77$  days. The female was larger than male.

### LEAF BEETLES OF GENUS *CHAETOCNEMA* STEPHENS, 1831 FROM SINDH, PAKISTAN

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Leaf beetles belong to the family Chrysomelidae is one of the largest families of coleopteran. Family comprises 19 subfamilies, with approximately 50,000 members within more than 2000 genera. Genus Chaetocnema Stephens, 1831 belong to the subfamily Galerucinae and tribe Alticini, they are known to feed on families; Poaceae, Cyperaceae, Chenopodiaceae, Convolvulaceae, Combretaceae, Polygonaceae, and Malvaceae, several of them are pests of commercial crops. They are characterized by having pronotum with no special features, elytra with confused punctures, middle and hind tibiae with a blunt projection or tooth in the apical half, followed by an emargination. For identification up to the species level often requires studying male genitalia. From Pakistan no detailed work on chrysomelidae is carried, hence keeping in view the importance of the family in agriculture, present work is proposed. For present studies beetles were collected from various localities of Sindh, Pakistan. Specimens were collected through traditional hand net, pooter and on light traps, all standard protocol methods for the collection were followed. Images for identification were captured with the help of DSLR camera fitted on rail, several images were taken at various depth of fields, further were staked and processed on software control zp. The specimens were identified using available keys. Map was prepared on google earth online. The male genitalia was dissected by removing and heating the pygophore in 10% KOH for 25-30 minutes, the line drawing was prepared with camera lucida and further converted on computer with Adobe illustrator CS6. During present studies 03 species of genus Chaetocnema were recorded including; Chaetocnema belli Jacoby, 1904, Chaetocnema concinnicollis (Baly, 1874) and Chaetocnema pusaensis (Maulik, 1926), all species were recorded from Sindh Province and several from commercial crops. The audience will have incite to habitus images, genitalia and distribution of species.

### TAXONOMIC STUDY OF BUMBLE BEES (HYMENOPTERA, APIDAE) FROM DISTRICT SHIGAR, GILGIT BALTISTAN

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The Bombini are a tribe of large bristly apid bees which feed on pollen or nectar. The tribe contains a single living genus, *Bombus*, the bumblebees, and some extinct genera such as *Calyptapis* and *Oligobombus*. The tribe was described by Pierre André Latreille in 1802. The genus *Bombus* Latreille, 1802 includes around 260 species of eusocial bees, mostly distributed in temperate and cold regions of the northern hemisphere. Bumble bees plays an important role in pollination and environmental indicators, identification of these bumble bees is also an important as different species play various roles in ecosystem. For the present studies bumble bee were collected from district Shigar. Further examination and identification were carried out at insect systematic laboratory, department of Entomology, Sindh Agriculture University Tandojam. During present study 8 species are recorded under single genus *Bombus*, from the collection site. Species including; *Bombus asiaticus* (Morawitz, 1875), *Bombus rupestris* 

(Fabricius, 1793), Bombus melanurus (Lepeletier, 1835), Bombus semenovianus (Skorikov, 1914), Bombus tunicatus (Smith, 1852), Bombus ferganicus (Radoszkowski, 1893), Bombus lucorum (Linnaeus, 1761) and Bombus rufofasciatus (Smith, 1852). The bees were collected from different host like Euphorbia wallichia, Cirsium arvense, Carduus edelbergii, Lavatera cashmeriana, Cirsium arvense, Melilotus officinale, Taraxacum officinale, Ipomea tricolor, Helianthus annuus, Digitalis purpurea, Zinnia elegans and Fagopyrum esculentum.

### RECOGNITION OF INSECT PESTS OF CHICKPEA (CICER ARIETINUM L.) AT TANDOJAM, PAKISTAN

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For present studies insect pests were collected from Chickpea crop at Pulse department (Oil Seed Section), Tandojam. Further examination and identification were carried out at insect systematic laboratory, department of Entomology, Sindh Agriculture University Tandojam. In present study total 90 specimens of class Insecta were collected from chickpea crop oilseed section at Sindh Agriculture University Tandojam, the study revealed the occurrence of various 08 species under 3 insect orders; Lepidoptera, Linnaeus, 1758, Orthoptera, Latreille, 1793 and Hemiptera Linnaeus, 1758. Lepidoptera revealed 3 records of species including; Helicoverpa armigera (Hübner, 1808) under family Noctuidae, Latreille, 1809 and subfamily Heliothinae, Boisduval, 1828; Danaus chrysippus (Linnaeus, 1758) under family Nymphalidae, Rafinesque, 1815 and subfamily Danainae, Boisduval 1833; Catopsilia pomona Fabricius 1775 under family Gryllidae Laicharting, 1781 and subfamily Gryllinae Laicharting, 1781. Orthoptera discovered with three species record including; Gryllus bimaculatus De Geer, 1773 under family Gryllidae Laicharting, 1781 and subfamily Gryllinae Laicharting, 1781; Gryllotalpa africana (Palisot de Beauvois, 1805) under family Gryllotalpidae Saussure, 1870; Chrotogonus trachypterus trachypterus (Blanchard, 1836) under family Pyrgomorphidae Brunner von Wattenwyl, 1874 and subfamily Pyrgomorphinae Brunner von Wattenwyl, 1874. Hemiptera Spilostethus hospes (Fabricius, 1794) under family Lygaeidae Schilling, 1829 and subfamily Lygaeinae Schilling, 1829; Acyrthosiphon pisum (Harris, 1776) under family Aphididae Latreille, 1802 and subfamily Aphidinae Latreille, 1802.

### AMPHICYON FROM THE CHINJI FORMATION OF LOWER SIWALIKS NORTHERN PAKISTAN

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New material of *Amphicyon* were recovered from the Chinji Formation (Lower Siwaliks) of Kanhatti village, Khushab district, Punjab, Pakistan. These specimens testify the species in the Lower Siwaliks after Lydekker (1884) and Pilgrim (1932). Amphicyonidae are very common in Carnivora from Early to Middle Miocene of Eurasia, North America and Africa. Referred specimen belongs to *Amphicyon palaeindicus* which is reported from the Chinji Formation outcrops of Khushab. This carnivoran species prey upon the smaller herbivores and other mammals at that time.

### NEW RECORD FOR TWO DIGENEAN TREMATODES OF FAMILY NOTOCOTYLIDAE FROM POCHARDS (ANATIDAE: AYTHYINAE) IN SINDH PAKISTAN

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During faunestic studies of trematodes of Anatid water fowls of local water bodies in Sindh Pakistan, a total 42 trematodes of family Notocotylidae including 26 specimens of genus *Notocotylus* were recovered from *Aythya nyroca* and 16 specimen of genus *Catatropis* were recovered from intestines of *Aythya fuligula*. A total of 38 hosts were examined in which 27 birds were found infected with went positive, specimen were further processed by following the method given by Schmidt and Garcia. On the basis of shape and size of the body, and other major characteristics these trematodes is identified as such *Notocotylus attenutus* (Rudolphi, 1809) and *Catatropis pakistanensis* Schuster and Wibbelt, 2012. *Notocotylus attenutus* and *Catatropis pakistanensis* is reported for the first time from pochards from Pakistan, however, these species are usually recovered from the anatid birds. Therefore the present trematode has been put as new host and new locality record at world level.

### DIVERSITY OF CASH CROPS POLLINATORS (HYMENOPTERA: SPP.) IN DESERT AREAS OF DISTRICT BHAKKAR AND LAYYAH

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The hymenopterans are considered most effective pollinators of various cash crops including maize, millet, wheat and cotton. As in agro ecosystem of desert areas large numbers of hymenopteran pollinators are found which need to be explored. So, keeping in view the importance of pollinator's present study was conducted to find out diversity, percentage occurrence and distribution patterns of the hymenopteran bees and wasps from district Layyah and Bhakkar. In each district, eight hotspots with quadruplicates were selected to observe pollinators population. The results reveal that 14 genera with 7 families were found on five cash crops. The recent findings indicate the honey bee (Apidae) as well as non-Apis bees (Vespidae) with less significant number than *Apis* bees. The community analyses of *Apis* and *Polistes* bees on cotton crops were dominant with AF=100% in both districts while *Scelipheron* and *Andrena* with AF=75% were observed in significant frequency in Layyah as compared to Bhakkar. From mustard populations Apis bees were dominant in both areas with AF=100% in Layyah and AF=87.5 from Bhakkar. Therefore, no significant difference was recorded among hymenopteran pollinators population from both districts but Shamon index indicated maximum diversity of pollinator on wheat and less diversity on millet in Bhakkar and on maize in Layyah. So the preliminary studies provide a base to document the total diversity and individual role of these pollinators in agro ecosystem and to develop policies to protect these pollinators and their habitat.

### BIODIVERSITY OF GROUND BEETLES (COLOEPTERA: CARABIDAE) IN UPPER SINDH PLAINS, PAKISTAN

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Ecological diversity of Carabid beetles was conducted in plains of upper Sindh for the first-time during August 2018-March 2019 among different habitats. The study was carried out at five sampling stations and adults were

trapped with the help of light trap, pitfall trap and hand-picking methods. A total 783 specimens of six species were collected, the maximum number of specimens and species was found in muddy soil and agricultural field at locality Nausheroferoz, least number was found in arid and bushy field. The month wise population showed that august is most abundant whereas declined in the month of February and cultivated habitat influenced the population dynamics. Biodiversity was calculated using Simpson diversity index, the sites 1 and 5 were with great numerical value of evenness and diversity i-e (E, 0.91 and D, 5.5) that reveals rarity and commonness of species in ecosystem. The whole dominant structure of Carabid complex revealed two most dominant species are Calosoma auropunctuam Herbst 1874 and Nesambloyps oreobious Broun 1893.

### PRELIMINARY OBSERVATIONS ON MICROPLASTICS FROM TWO SELECTED PICNIC SPOTS AT RIVER KABUL AND RIVER SWAT. PAKISTAN

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Microplastics (MPs) is the term used often for the size of plastic particles lesser than 5mm. Microplastics are extremely hazardous as they are ingested by invertebrates which form a large component of fish diets thus bioaccumulate and influences the higher trophic levels eventually reaching human beings. Aim of the present study was to assess the presence of microplastics in different waterbodies of Khyber Pakhtunkhwa. Sand samples from four stations located at River Swat (KS1, KS2) and River Kabul (SS1, SS2) were taken, processed by density separation method using NaCl, filtered using 38um filter paper and then analyzed under Stereomicroscope (40X). Results were as follows; (KS1 = 800 MP Particles kg<sup>-1</sup>, KS2 = 1100 MP Particles kg<sup>-1</sup>, SS1 = 800 MP Particles kg<sup>-1</sup>, SS2 = 900 MP Particles kg<sup>-1</sup>). Mostly microfibres were found showing the more use of clothing materials made of artificial fibers and its disposal via washing of clothes. MPs are hazardous for aquatic life as aquatic fauna accidently ingest them as food or with food. It was concluded that River Swat is a bit more polluted than River Kabul. It is recommended that use of synthetic fiber materials should be minimized in the textile industry and laws should be made to lessen the use of plastic materials. Similarly Government should focus on reducing the inputs of MPs to freshwater river systems by introducing the sewage management system with the latest filtration technologies which is an efficient way of minimizing MPs in disposed water.

### NEW RECORDS OF PHTHIRAPTERAN ECTOPARASITES FOUND ON WILD QUAILS (GALLIFORMES: PHASIANIDAE) OF SINDH, PAKISTAN

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The quail is an important recreational bird of Sindh Pakistan. There are 08 species of *Coturnix* in the world, amongst which 04 are found in Pakistan, affected from different types of parasitic diseases among which lice infestation is considered as the most prevalent infestation. Quail has prominent importance in health and nutrition culture like; diseases which are treated by using quail eggs are asthma, Tuberculosis, and diabetes and remove various types of stones. Such diseases are popularly treated by using quail eggs in China. Its meat is very rich in micronutrients, absolutely allergy free with low cholesterol values, nutritious, tender and tasty. This is the first ever study on the ectoparasites of *Coturnix* in Sindh, Pakistan. The study told the presence of ectoparasites on *Coturnix* from the different districts of Sindh. All the collected birds were inspected for the ectoparasites. The prevalence of ectoparasites on all species of *Coturnix* was found 100% from all the collected 100 birds. The collected lice were preserved in 70% ethanol, prior to slide preparation using KOH and passing from ethanol series. The species were mounted in Canada balsam using standard procedure for the identification of species ahead. During this study, lice were collected from 03 host species viz. *C. japonica*, *C. coromandelica* and *C. coturnix* of Sindh. The host birds were

abundantly found infested by 05 genera of 02 families' viz. Family Menoponidae (four species of Amblycera) and Family Philopteridae (three species of Ischnocera), and all were recorded exclusively from the Sindh region ever in history, making the initial country records while as Menacanthus pallidulus (Neumann, 1912) Menacanthus abdominalis (Piaget, 1880), Columbicola columbae (Linnaeus, 1758), Colpocephalum turbinatum (Denny, 1842) and Colpocephalum barbus sp.n., have also made new host records too. Menacanthus pallidulus (Neumann, 1912) was found with highest prevalence and intensity while as Colpocephalum barbus sp.n. was investigated with lowest prevalence. Hence, there were seven chewing lice species reported from Sindh, Pakistan. It is the first chewing lice study on Coturnix species new host record in the world hence, the new record has been found from Sindh, Pakistan.

### SEASONAL EXISTENCE OF FOLIAGE INSECTS IN ORNAMENTAL FLOWERS FIELD

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Biodiversity is the variety of species at all level of organization either genetic, molecular or ecosystem level. It describes the services to human societies, richness of species and efficiency of ecosystem, therefore proves profitable. This research work was done to find out the foliage insects diversity among the jasmine and muraya Crops under the climate of district Faisalabad. From the whole 2534 noted insects from both crops and tallest abundance was documented for Murraya crops. Population means per samples was also calculated along with standard deviation (SD) In in the Murraya crops largest no of insects species with tallest mean and standard deviation noted from  $7^{th}$  samples  $15.67 \pm 188(50.09)$ . After calculation, From Murraya crops overall 1406 insects with different relative abundance was noted and largest relativity 4.26% (N = 60) was logged for *Lucilia sericata*. ANOVA was made. After the completion of the analysis, it was showed that population mean of noted taxa among both territories (Murraya and Jasmine) non-significant. Presently trophic level arthropods inhibiting in fragmented landscape viz. Murraya and Jasmine was accessed from different season of these crops. Afterwards of the completion of the analyses, it was observed that in overall strength, that fauna was existed non-significantly in both crops (t-value = 0.19; P-value=0.0477) for one tail and P-value 0.059 for two tail. It was observed that population mean of noted taxa among both crops (Murraya and jasmine) showed non-significance results (F=0.01; P=0.0009946). It was assured that structural community and taxa composition were showed significant results among all crops (F=0.01; P <0.0009946).

# IMPACT OF DIFFERENT LIGHT INTENSITIES ON TH DISTRIBUTION FREQUENCY OF NOCTURNAL INSECTS

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Insect diversity contains a massive part of biodiversity around the globe. Among them, nocturnal invertebrates are more energetic at nighttime than day. Artificial light fascinates numerous species of nocturnal insects and light traps provide momentous indication for the nocturnal insect diversity that how much attraction they have towards light of different wavelengths. Overall 35278 specimens were recorded from lights of different watts, and maximum population was observed from 500 W light 48.93% (N = 17261) and least population was noted at 25 W light 3.49% (N = 1233). After accomplishment of research, total 91 species from 25W, 105 species from 100W, 171 species from 200 W and 162 species were recorded from 500W. Biomass from 25W was (2.72±1.63), from 100W, (1.93±0.69), from 200W (25.48±11.39). From 25W light, *Nysius raphanus* was noted with high number and relative abundance 26.52% (N = 327), 100W light, *Aphis caliginosa* was noted as an extra participant with relative abundance 25.69% (N = 1046), 200W light, *Aphis glycines* 25.87% (N = 3288) and 500W light *Macrosiphum rosae* 33.08% (N = 5710)

was documented. Overall 102 documented families, 50, 56, 79 and 71 families were noted from 25, 100, 200 and 500W light respectively. In different lights frequencies viz. 25, 100, 200 and 500W, all the 11 orders were noted. From 25W, 100W, 200W and 500W lights Hemiptera was documented for high relative abundance 58.39% (N = 720), 84.92% (N = 3458), 84.63% (N = 10758) and 82.20% (N = 14189) respectively. Diversity was documented max. (2.69) for 500W light.  $H_{max}$  was recorded highest (5.14) from 200W while max. evenness was noted for 25W.

## EXPLORING THE MOTH (LEPIDOPTERA) FAUNA OF DISTRICT BUNER KP, PAKISTAN

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The current preliminary survey of moth (Lepidoptera) was conducted from September 2018 to September 2019 in different localities of District Buner, Khyber Pakhtunkhwa, Pakistan. It was the first attempt to explore and catalogue the moth fauna of this area. In current investigation we focused on moth diversity assessment in nineteen different localities of District Buner. Each locality was visited for collection. In current exploration a total of 563 specimens of moth were collected. Entomological survey revealed that the area is more diversified with an annotated checklist of 28 species belonging to 25 genera under 8 families. Species calculation on family basis as Family Erebidae 8 species, Family Sphingidae 6 species, Family Noctuidae 6 species, Family Geometridae 2 species, Family Crambidae 2 species, Family Notodontidae 2 species, Family Lasiocampidae 1 species and Family Bombycidae 1 species. During collection sampling the most abundant species was *Aloa lactinea* (Cramer 1777), while the rare species was *Olene mendosa* (Hubner 1823). The reported species were also analyzed for morphometric measurement i.e. body width and body length.

# NEW RECORD OF *DIPLOTRIAENA SHAHRI* N.SP. (NEMATODE: FILARIIDAE) FROM JUNGLE MYNA (*ACRIDOTHERES FUSCUS*) WAGLER; 1827 (PASSERIFORMES: STURNIDAE) IN DISTRICT LARKANA, SINDH, PAKISTAN

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A new nematode *Diplotriaena shahri* n.sp is recorded from the body cavity of Jungle Myna *Acridotheres tristis* of District Larkana, Sindh, Pakistan. In all, 25 nematodes (3) were recorded. Present nematode reflects diversification from their congeners in the following charaters viz: body measurement; shape; size and shape of trident; size and shape of spicules. On the basis of such morpho-metrical changes this species; *Diplotriaena Shahri* may be treated as a new species. This new species is dedicated in the honor and name of Younger elder brother Mr. Shahryaar Khan Soomro.

# IMPACT OF EXTRACELLULAR MATRIX ON THE EXPRESSION OF EPITHELIAL TO MESENCHYMAL TRANSITION MARKERS: AN IN-DEPTH STUDY OF CROSSTALK BETWEEN TUMOR MICROENVIRONMENT AND CELL ADHESION MOLECULES

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Extracellular material is a dynamic component of the tumor microenvironment. The extracellular material mostly contains collagen, proteoglycans and some glycoproteins. Each of these components serves a special role.

Some components are associated with high tumorigenesis. For example, collagen alone being the most abundant in ECM and diverse in nature, can promote cancer metastasis and enhance proliferation by upregulation integrin components. Some other ECM components have been thought to inhibit tumor metastasis, For example, elastin is abundantly found in skin, bones and vessels where tumors rarely undergo metastasis, except for the lung which is highly metastatic site with high elastin. It has been found that the soluble form of elastin has an inhibitory role on metastasis. We have used two major ECM components Collagen (Gelatin 50 Bloom Sigma®) and Elastin (Sigma®) as a coating material for cancer cell attachment. We studied the expression level of E-cadherin, N-cadherin, Integrin  $\alpha$ 5, Integrin  $\beta$ 1, Integrin  $\beta$ 2, Snail, Slug, ZEB1 and ZEB2 on coated plates and its comparison to non-coated and tissue culture-treated plates (SPL®) for Hela, HepG2, MDA-MB231 and MCF7 cells through real-time PCR technique and microscopic approaches for different time points up to 3 days. Our study suggested that exogenous expression of these coating material has implication in cancer progression and these can be targeted to localize the tumor progression *in-vivo*.

## ECOLOGICAL ASSESSMENT OF HERPETOFAUNAL DIVERSITY IN QAZINAG FOREST RANGE, DISTRICT JHELUM VALLEY, AZAD JAMMU AND KASHMIR

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Herpetofauna have important role in the local ecology, cultural and aesthetic values of the local communities. Due to lack of scientific information their proper management cannot be prioritized and most of biological diversity of herpetofauna is unexplored in Azad Jammu and Kashmir (AJK). This study is designed to explore the diversity of herpetofauna (amphibian and reptiles) along with focusing on their major ecological attributes in the Oazinag Forest Range. Five major habitats in twenty different sites were surveyed to collect the field data during August 2018 to August 2019. Visual encounter surveys were performed during dawn and dusk hours a well as during nights (for nocturnal species) in different potential habitats. A total of 827 specimens belonging to 10 families, 22 genera and 27 species of herpetofauna were recorded. This included five species of order Anura (Amphibia) and 22 species of order Squamata (Reptilia). Among these, 16 species were locally considered as rare, while remaining 07 and 04 species were abundant and common respectively. However, according to IUCN (2019), 18 species were classified as Not Evaluated (NE) and while other species were (n=7) Least Concern (LC) globally. The highest species richness was recorded in Nardajian (n=25), followed by Khatarnaar and Chham (n=24), Pandu (n=18) and Tararan (n=15). The abundance and relative abundance were recorded in Khatarnaar (n=285, 0.34), while the least in Pandu (n=100, 0.12). The highest species richness was observed in grasslands (n=26) and the lowest in water bodies (n=14). However, the abundance was the highest near human settlements (n=233) followed by grasslands (n=209), water bodies (n=153), agricultural fields (n=131) and forests (n=101). The diversity index was highest in forest habitats (H'=2.503) followed by agricultural fields (H'=2.482), human settlements (H'=2.136), and grasslands (H'=1.983). The species richness and abundance were higher in post-monsoon (n=27, 418) than the pre-monsoon season (n=25, 409) with the diversity indices of 1.98 and 2.35 respectively. Several factors were found influencing the diversity of herpetofauna in different localities. Habitat loss, fire and human killings were the most severe threats to the majority of herpetofaunal species in the study area. Unawareness and lack of proper education is a serious issue in Qazinag Forest Range and most of the species killing were due to lack of awareness. This study provides baseline information about the herpetofauna and will help in formulating the future conservation management strategies in the study area.

# ANISODON SALINUS REMAINS FROM THE CHINJI FORMATION OF THE PAKISTAN SIWALIKS

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Chalicotheres are rare throughout the world and the same is applicable to these clawed herbivores in the Siwaliks. Eight new dental remains have been described from four localities of the Chinji Formation (14-11 Ma), Lower Siwaliks of Pakistan. The new specimens are attributed to *Anisodon salinus*, previously known as *Chalicotherium salinum*. The specimens are significant not only because of their rarity but also because of what they add to information about the morphological and metrical values of this species. The phylogenetic status of the Siwalik species is still debated and still requires a detailed analysis.

### TREMATODES OF HYDERABAD, JAMSHORO AND NAUSHAHRO FEROZE DISTRICTS, SINDH, PAKISTAN

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During the survey of helminth parasite 26 Egretta garzetta, 22 Sternula albifrons, 05 Himantopus himantopus, 14 Bubulcus ibis, 20 Ardeola grayii were shot down from the Hyderabad, Jamshoro, Naushahro feroze Districts, Sindh, Pakistan. Total 254 specimens of trematodes were recovered from small intestine of hosts. The specimens were mounted according to standard procedure and identified as belonging to genus Uvitellina Witenberg, 1923; Cotylurus Rudolphi, 1808; Apatemon Szidat, 1927; Episthmium Lune, 1909; Echinostoma Rudolphi, 1809; Heterotestophyes Leonove, 1957; Knipowitchiatrema Issait Schikow, 1927 and Stictodora Looss, 1899. Uvitellina megacaecatum sp.n reported from Himantopus himantopus; Cotylurus cornutus (Rudolphi, 1808, Szidat, 1927) Bhutta and khan, 1975 and Apatemon sp. both recovered from Bubulcus ibis; Echinostoma rafiae sp.n; E. garzetti sp.n; Episthmium jamshorensis sp.n; E. sindhensis sp.n; E. bilqeesae were reported from Egretta garzetta; Heterotestophyes heckmanni sp.n; H.gibsoni sp.n; H.jonesae sp.n; Knipowitchiatrema pakistanesis sp.n; K.sternulai sp.n were recovered from Sternula albifrons and Stictodora alykhani sp.n from Ardeola grayii.

# SYSTEMATICS OF CICADELLINE LEAFHOPPERS (HOMOPTERA; CICADELLIDAE) OF PAKISTAN. A REVIEW

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Cicadellidae is considered as the most diverse family of Homoptera, consisting of about 22,000 described species in over forty subfamilies and above 35,000 species of infraorder Cicadomorpha. Leafhoppers are the soft bodied, minute insects of the family Cicadellidae that suck plant sap from grass, shrubs or trees. Cicadelline leafhoppers are well studied leafhoppers, because they are pests of many important cultivated crops and vectors of plant pathogens. In Pakistan, some of the major sub families of economical importance are studied from previous century and the studies are ongoing until now. Sub family, Typhlocybinae comprehensively studied after the mid twentieth century in province of Punjab as well as Sindh. Another sub family Deltocephalinae is well described in

Sindh, Tandojam. Taxonomic research on sub family Cicadellinae is little known in Pakistan. In this review, we have concluded that there is a huge work remains to be done on leafhoppers; ongoing studies may reveal even more significant importance of Cicadellidae.

#### TAXONOMIC STUDIES OF THRIPS ON COTTON IN DISTRICT MUZAFFAR GARH

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Cotton is the major fibre crop in Pakistan. Pakistan is at 5<sup>th</sup> position among cotton exporting countries and third in cotton consuming countries in the world. Its contribution in country's GDP is about 10%. There are many reasons for lower production of this crop and attack of insect pests is the major one. Many species of insects and mites are reported from Pakistan in cotton. Among the sucking insect pests of cotton, Thrips are fairly destructive pest of many crops including cotton. Thrips are small soft bodied insects which belong to the order Thysanoptera. *Thrips tabaci* Lindeman has become regular pest of cotton in the Punjab Pakistan. In cotton, thrips are reported to cause the loss in yield up to 50%. Different species are reported in literature on cotton in Pakistan, due to which Farmers are facing difficulties in controlling this pest, may be due to some cryptic species. That is the reason due to which control measures against this pest are not useful. So the present study is being devised to check species of this pest on crop. My work is under study and by following taxonomic key still I found that Specie infesting cotton is *Thrips tabaci*. Further in future may be any specie reported from my research.

## STUDY ON TAXONOMIC STATUS OF GROUND CRICKETS (NEMOBINAE) FROM DISTRICT KHAIRPUR

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The insects belonging to subfamily Nemobinae are commonly known as ground crickets. They are smaller in size, somewhat flattened in structure. Fastigium of vertex usually rounded. Ocelli smaller and triangular in shape. Pronotum subquadrate. Hind femora with slight hairs and long spines. Ovipositor needle shape. Cerci elongated slightly cylindrical. Extensive surveys were carried out to collect ground crickets from district Khairpur. A total of 193 specimens were captured and were identified into single sub-family Nemobinae with 05 genera and 07 species i-e: *Pteronemobius indicus* (Walker, 1869), *Speonemobius decolyi* (Chopard, 1969), *Pteronemobius concolar* (Walker, 1871), *Nemobius fasciatus* (De Geer, 1773), *Allonemobius socius* (Scudder, 1877), *Allonemobius fasciatus* (De Geer, 1773) and *Eunemobius carolinus* (Scudder, 1877). Of which *Allonemobius socius* (Scudder, 1877), *Allonemobius fasciatus* (De Geer, 1773) are reported for the first time from Sindh Province. While remaining 05 species are first time reported from district Khairpur Mirs. Beside this, highest population of *Pteronemobius concolar* (Walker, 1871) with 30% followed by *Pteronemobius indicus* (Walker, 1869) with 23% and lowest population of *Eunemobius carolinus* (Scudder, 1877) with 3% followed by *Allonemobius socius* (Scudder, 1877) with 7% was recorded respectively. In addition to this, description of species along with habitus images are provided. Hopefully, this study will be a base line for future researcher.

### NEW RECORD OF *RHOMBODERELLA SCUTATA*, BOLIVER, 1889 (MANTIDAE: MANTINAE) FROM SINDH PAKISTAN

#### Kashif Ali Shar and Waheed Ali Panhwar\*

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Praying mantises belongs to Superfamily Mantodea feed on various insects and are regarded as predators. Due to their predatory nature mantises play an important role for the control of pest. Mantids are very unique and among

the most easily recognized of all insects. At the present detailed surveys were conducted from several talukas of district Naushahro feroze. *Rhomboderella scutata*, Boliver, 1889 is reported as new regional record for Pakistan. Additionally, photographs are provided for the first time. Finding of present study will contribute to the biodiversity of Pakistan.

#### IMPACT OF LEAD ON FRUIT FLIES IN MULTAN, PUNJAB PAKISTAN

### Immad Anwar\*, Faheem Aslam, Muhammad Ramzan, Masood Maqbool, Ghulam Murtaza and Syed Haroon Masood Bokhari

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The heavy metals like lead are important toxic and dangerous material which directly or indirectly contaminates the foods, soil water, and air, accumulate in insect body like fruit fly, causes organogenesis, sperm abnormalities, damage to nervous system, morphogenesis syndromes and miscarriage. The current study was carried out to check the lead effect- ion on various developmental stages of fruit fly at Muhammad Nawaz Shareef University of Agriculture, Multan in 2018. For this purpose, six pairs of flies were collected from field and added to culture consist different concentrations of lead-ion for mating and egg lying. During the present study, the transformation rate of larva-pupa, pupa-adult and adult-egg lying, morphological changes and time required for each stage were studied. The different morphological changes like elongated wings, de-shaped wings, elongated and folded legs were observed in current study. The study resulted that larval and pupation period was increased with 25-300 mg/L culture- medium lead- ion, while alteration of larvae to pupa and pupa to adult reduced. The lead has negative effect on egg hatching period. The larval, pupal and adult growth rate was reduced with metal concentration. The study concluded that lead interfere with enzymes or hormones that evolved in metamorphosis and ATP synthesis.

### A NEW RECORD OF TREMATODE FROM COMMON TEAL ANAS CRECCA L. (ANSERIFORMES: ANATIDAE) FROM MAJOR WATER BODIES OF SINDH, PAKISTAN

#### Shaila Khaskheli\* and Saima Naz

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In the present study of common teal *Anas crecca* (Anseriformes: Anatidae), a total of 73 hosts were collected from Manchar Lake of district Jamshoro and Keenjhar Lake of district Thatta and were examined thoroughly for the presence of helminthic infection. The infection of cestodes was observed maximum (45%) along with all helminths in the host species. Additionally there is no more work has been done on trematodes from this host from Pakistan, which revealed a new record of trematode, *Echinoparyphium recurvatum* (Linstow, 1873) from *Anas crecca* in Sindh region making a new host and new locality record.

## A MODEL FOR SMALL SCALE DUCKWEED BASED, MANAGED ECOSYSTEM FOR DISTRICT CHINIOT, PUNJAB, PAKISTAN

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Duckweed is an aquatic floating plant with 1-3 leaves. It is highly nutritious containing 40-45% protein, 15-30% fiber, 5% fat and 27-34% starch and hence can serve as animals feed and fertilizer for vegetation. Duckweed can also absorb toxic heavy metals and can help to conserve water. In district Chiniot, waste water is frequently seen around residential localities forming stagnant ponds with excessive duckweed growth. The duckweed utilization

model aims to represent stages of water purification and conservation through duckweed. The municipal waste water containing high levels of heavy metals can be purified through absorption by duckweed. The heavy metal free water can be channelized to the duckweed production pond where fresh seed stock of duckweed can be added. This duckweed will supply animal feed and fertilizer while purified water can be used to culture fisheries and supply water to vegetation. Thus, we propose that duckweed can be effectively used as a fertilizer for vegetation, as fish, cattle and chick feed while enabling waste water purification and conservation. It is not only an important tool for bioremediation with its water purifying properties but also an excellent mosquito repellant.

### ROLE OF TERMINALIA ARJUNA IN MEDICAL FIELD AND ITS BENEFICIAL ASPECTS WITH REFERENCE TO INSECTICIDES. A REVIEW

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Mankind and medicinal plants have a strong historic association. In the past they were only used for the healing of wounds but now with the evolution of awareness and advancement, pharmacists have become able to tackle new diseases by extracting multiple phytopharmacological compounds to respond to the challenges of medical field in better way. *Terminalia arjuna* (Combretaceae) has been reported beneficial for cardiovascular patients suffering from Angina (chest pain) and ischemic heart disease. Its bark extract has Antioxidants (flavones, tannins, lateilin, oligomeric proanthocyanidins), Acids (arjunic acids, tomentosic acid, gallic acid, ellagic acid, terminic acids), Glycosides (arjunetin, arjunine, arjunosides) and minerals that exhibit anti-ischemic properties. Besides, it is also known as liver tonic, lowers blood cholesterol level and exhibits antibacterial, antifungal, insecticides and anti diabetic properties. Arjuna is also used in the treatment of sexually transmitted disease gonorrhea because of its specific activity against bacterium Neisseria gonorrhoeae. Its intrinsic compounds render this tree as growth inhibitor and feeding-deterrent for many insect species. In this study, *Terminalia arjuna* is a great blessing of God, steps should be taken to commercialize its products for the uses of masses.

### PHYSICO-MORPHIC TRAITS OF COTTON IN REGULATING POPULATION DENSITY OF SUCKING PEST COMPLEX. A REVIEW

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Cotton is highly cash crop of Pakistan which contribute significant role in the economics and major source of foreign exchange earnings. Pakistan is multi insect complex system in case of cotton; it is vulnerable to attack from a variety of chewing and sucking pests. It is difficult to manage sucking pest because of its negative correlation with chewing pests. The cotton traits like leaf and boll characteristics including: leaf color, leaf length, leaf lamina thickness and trichome density and trichome length, which are positively correlated with *Bimisia tabacci*. Whiteflies are less attracted to the light green plant leaves as compared to the leaves with bright green color. Plants physiomorphic characteristics number of hairs on midrib, lamina and vein are negatively correlated with Amrasca biguttula biguttula, while thickness of leaf lamina and gossypol glands on midrib is positively correlated. In this study, we suggest that make a genome, which would act as a feeding deterrent for sucking insect pests.

## COBALT TOXICITY TO THE ECONOMICALLY IMPORTANT CARNIVOROUS FISH WALLAGO ATTU (MALLI)

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Increasing metal concentrations in freshwaters of Pakistan may cause serious health and survival issues for the carnivorous fish. Therefore, the research work was conducted to assess the toxicity of commonly found aquatic contaminant (cobalt) to one of the significant carnivorous fish species found in riverine systems of Pakistan i.e. *Wallago attu* known as malli. The toxicity trials were performed under constant laboratory conditions. During the trials, 96 h acute toxicity of cobalt and its bioaccumulation patterns were determined for W. attu of 150 mm total length. The fish mortality data were recorded and analyzed statistically by employing Probit analysis method to determine 96 h LC<sub>50</sub> and lethal concentrations of cobalt for W. attu. The 96 h LC<sub>50</sub> and lethal concentration values of cobalt computed for W. attu were  $98.11\pm2.72$  and  $133.84\pm5.12$  mgL<sup>-1</sup>, respectively. At 96 h LC<sub>50</sub> and lethal concentrations exposure, metal accumulation in skin, gills and gut of the fish was also analyzed by using atomic absorption spectrophotometer. Statistically highly significant differences at p<0.01 were recorded among the body tissues of fish to accumulate cobalt. At both 96 h LC<sub>50</sub> and lethal concentrations exposure, cobalt accumulation in body tissues of W. attu followed the order: gills > gut > skin.

#### REGIONAL TAXONOMIC STUDIES OF SPIDER'S FAUNA OF PAKISTAN: A REVIEW

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Arachnids are a diverse group of arthropods including spiders, which plays a significant role in ecology of agroecosystems as predators of many insect pests of cultivated crops. In wild ecosystems, they are also a part of food chain. Spiders consist of 42751 species of 3859 genera described species belonging to different families worldwide. Pakistan is gifted with diverse habitats, which provides a huge diversity of spiders. In past decade, there are many regional taxonomic studies conducted on spider's fauna in Pakistan. Regional taxonomic studies are reviewed and listed in this paper along with a checklist of different species reported in Pakistan. There is little work known in Pakistan related to medicinal, aesthetic and in pollinator aspect of spiders. In future, this article will help to explore new fields of scientific research on spiders.

## NEW FOSSIL COLLECTION OF *HIPPOHYUS SIVALENSIS* (ARTIODACTYLA: SUIDAE: SUINAE) FROM LATE MIOCENE TO PLIOCENE OF SIWALIKS OF PAKISTAN

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Hippohyus sivalensis is a common suid of late Miocene to Pliocene rocks of Tatrot/Hasnot area of Pakistan. The molar resemblance with equids indicate their grazing feeding habits. This species migrated to Potwar land when grassland established there. It has typical suine characters with hypsodont dentition. The described material consists of isolated molars. This discovery will provide a new insight to understand the diversity and geographic distribution of Siwalik Suids.

### PREVALENCE OF HUMAN MALARIA PLASMODIUM AT TEHSIL MATTA DISTRICT SWAT

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Malaria is one of the most important transfusion associated infections in many parts of the world, particularly the developing countries where it is endemic. This study was conducted to determine the prevalence human malaria in Tehsil Matta, Swat. A total of 4622 blood samples included male and female were collected from different labs of Tehsil Matta and were screened for the presence of malarial parasites in thick and thin smears using microscopy. Malaria positive samples were further conformed through diagnostic PCR. A total of 4622 samples 166 were found positive for malaria parasite. 99.40 % cases to *Plasmodium vivax* and 0.40% was *Plasmodium falciparum*. Majority of the patients belonged to age group 21 - 40 years. Infection was more common in males as compared to females (65.7%, 34.3%). Data also showed that the prevalence of malaria is more common in the month of August to September and less common in the month of January to March. Highly prevalence was found in Union council Matta Kharirai and UC Baidra while less prevalence in UC Beha. It is concluded from the present study that malaria infection is more common in tehsil Matta. Further research on this aspect using more sophisticated and advanced diagnostic. Techniques are required for proper assessment of actual situation and control of transmitted malaria in Pakistan.

### NANO-PARTICLES: AN ALTERNATE TECHNIQUE FOR MANAGEMENT OF DENGUE VECTORS; A REVIEW

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Mosquitoes transmit many diseases in human and animals, thus play important role in the environment for their pathogenicity. To reduce their risk to human and domesticated animals is a critical issue since times immemorial. Now a day, much emphasis is towards chemical insecticides for their control that lead to environmental pollution and resistance in target species. To mitigate these problems, alternate control measures like biopesticides, bio-control agents and genetically modified mosquitoes, are being adapted for suppression of their population. Among these, a comparatively new method is the use of nano-particles synthesized from plant extracts. Many plant extracts are used as aqueous solution for green synthesis of silver nano-particles (AgNPs) against larvae of *Aedes aegypti* and has been proved effective. Aqueous solutions of plant extract from *Chrysanthemum*, *Anisomeles indica*, *Melia azedarach* and *Artemisia vulgaris*. These are also eco-friendly and safe for other non-target species especially pollinators and predators. *Not only plants extracts but a number of micro organisms such as bacteria* (Bacillus thuringiensis) *and fungus* (Beauveria bassiana) are used to synthesize *Cobalt nanoparticles* (*CoNPs*) *against* Aedes spp.

### EFFECTIVENESS OF BUMBLEBEE POLLINATION ON HYDROPONICALLY GROWN CHERRY TOMATOES

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Protected environment cultivation of tomatoes and other vegetables considered as costly crop cultivation. Lack of plant movement due to absence of wind in controlled environment affects badly proper pollination. The tomatoes

which are grown in greenhouse require supplemental pollination for fruit set. For this purpose, a study was conducted at Hydroponic Unit, MNS-UAM. Cherry tomato was grown in controlled environmental condition. A bumblebee (Bombus terrestris) box containing 150 workers was kept in unit to compare its pollination with vibration and self-pollination method. Different parameters including physical, biochemical and shelf life were evaluated and the results of this study revealed that bumblebee pollinated cherry tomato flower produced better fruits in terms of physical, biochemical and shelf life. Bumblebee visits were also evaluated and it was revealed Bumblebee visit 3 exhibited maximum fruit dimension, fruit weight, no. of seeds/fruit, weight of 100 seeds, acidity, Vitamin C and shelf life. While total soluble solid, pH and daily weight loss percentage was maximum in self-pollinated fruits. It is recommended that bumblebee is a good pollinator of cherry tomato grown in hydroponic unit and three visits of b bumblebee are enough to produce better cherry tomato fruit.

# CONSERVATION OF NATIVE SOLITARY BEES THROUGH THE SEASONAL ORNAMENTAL FLOWERING PLANTS

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Pollination has a significant importance in the reproduction of many crops and wild plants. It is performed by different pollinators. Bees are primary pollinators. Majority of the bees are solitary bees due to their living status. Population of the pollinator is decreasing due to loss of their habitat, diseases and unfavorable conditions. Ornamental flowering plants are commonly used to conserve the pollinators, to control erosion and to improve the environment in urban and sub urban areas. These are short term, high unit value crops and mostly cultivated in small acreages. The attractiveness of the ornamental flowers is 100 times greater in insects than in the humans. Some previous studies show that in family Asteraceae, *Tithonia rotunadifolia, Centaurea macrocephala* and *Dahlia pinnata* are valuable plants for bumble bees and solitary bees. *Lavandula* is the most common genus which is most attractive for bumble bee. Honey bees, solitary bees, bumblebees, hoverflies; butterflies and moths are most attracted pollinators. To conclude that it does not involve any gardening effort or extra cost to conserve the pollinator in garden.

#### PREVALENCE OF SUPERFICIAL HUMAN PATHOGENIC FUNGI IN, GILGIT

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Fungi are eukaryotic unicellular such as yeasts to multicellular organisms such as puffballs with cell wall composed of chitin. They are fundamental for life on earth in their roles as symbionts, e.g. in the form of mycorrhizae, insect symbionts, and lichens. The total number of eukaryotic species on Earth has recently been estimated at 8.7 million, with fungi making up approximately 7% (611,000 species) of this number .The fungal lineage is one of the three large eukaryotic lineages that dominate terrestrial ecosystems. They share a common ancestor with animals in the eukaryotic super group Opisthokonta. Fungi are of an ancient lineage and have a fossil record that extends back to the Devonian and Pre-Cambrian era. The earliest written record of fungi are not of the fungi themselves, but of their depredations. To the physician and poet Nicander [ca. 185 B.C.], fungi were 'the evil ferment of the earth; poisonous kinds originating from the breath of vipers. Mycology is that branch of biology where we studying about fungi. Fungi are multicellular eukaryotic fungi ranging from unicellular to multicellular fungi like yeasts. They are fundamental for life on earth in their roles as symbionts, e.g. in the form of mycorrhizae, insect symbionts, and lichens. Likewise fungi have its own significance regarding its beneficial to harmful effect ever on plants, animals or humans. Human pathogenic fungi is more prevalent in our environment due its consequences on

human's hiddnley that's why called as "Hidden killers". Medical Mycology, a study of fungal epidemiology, ecology, pathogenesis, diagnosis, prevention and treatment in human beings, is a newly recognized discipline of biomedical sciences, advancing rapidly. Fungal infections today are among the most difficult diseases to manage in humans. These diseases kill more than 1.5 million and affect over a billion people. Fungal diseases are a worldwide problem ranging from superficial infections easy to cure to more invasive life threatening infections that are much harder to diagnose and treat. Recent estimates suggest that invasive fungal infections cause at least as many deaths as malaria and tuberculosis. The burden of fungal disease continues to increase as the number of people with weakened immune system increase. They attack people with serious illness, and frequently jeopardize the success of the newest medical advances in cancer care, solid organ and hematopoietic stem cell transplantation, neonatal medicine, autoimmune disease therapies, trauma and intensive care, and sophisticated surgery. Gilgit city is the main city of GB, which is the main attraction area to people for its beauty and resource purposes like education sectors, job opportunities and health facilities. People use together for different purposes related to job, study or other work. This city is mainly surrounded by other towns like danyore, Shanglidaar and oshikhandass. There are 4 main hospitals found in Gilgit are; DHQ hospital, City hospital, GMC hospital and CMH hospital Gilgit. Along with these hospitals one newly build hospital consist of 30 beds known as Shanglidaar hospital located in between Muhammad Abad danyore and oshikhandass. According to my area of research 3 hospitals were chosen to collect human fungal skin samples are; DHQ hospital, City hospital and Shanglidaar hospital. Total of 30 patients were examined collectively from these hospitals, 14 fungal species (Cryptococcus Neoformans, Actinomyces Israelii, Ajellomyces, Candida albicans, Cladosporium, Coccidioides ssp, Cryptococcus Neoformans, Emmonsia ssp, Geotrichum Candidum, Lacazia Loboi, Microsporum audouinii, Microsporum canis, Malassezia Fur Fur and Trichophyton Rubrum). The identified ssp belonging to 8 families and 10 genera the highest number of ssp is from Tinea which is four in number of genus Trichophyton. Identification was done in KIU biology Lab.

# PHARMACOLOGICAL STUDY ON SPINY-TAILED LIZARD UROMASTIX HARDWICKII GRAY (REPTILIA, LACERTILIA, AGAMINAE)

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Due to great versatility in the responses of smooth muscle, every step regarding pharmacological aspects always provided new clues to the researchers. Various pharmacological studies have been made on mammalians and amphibians smooth muscle while only few studies reported on reptiles. Further, Uromastix smooth muscle was never been considered regarding the pharmacological studies of Caffeine (Caff), Nor-adrenaline (NA) and Acetylcholine (ACh). Therefore, the present study is designed to obtained the responses from Uromastix ileum under the influence of Caffeine, Nor-adrenaline & Acetylcholine & their comparison with each other. 2mM & 4mM conc. of Caffeine, Nor-adrenaline & Acetylcholine were used to observe the changes in basal tone of the ileum of Uromastix, using organ bath assembly along with the isometric transducer on oscillograph. It has been observed that higher Caffeine concentration (4mM) was responsible for significant decrease (P<0.025) in the basal tone when compared with the low concentration (2mM) of caffeine significant change was also observed when Nor-adrenaline alone compared with Caff in the presence of NA and NA in the presence of caffeine. Furthermore Caff alone was also responsible for significant (P<0.05) decreased in basal tone when compared with Caff in the presence of ACh. This study demonstrates inhibitory influence of Caff & NA on Uromastix ileum, being smooth muscle relaxant, which is not yet reported in this animal model. It is suggested that inhibitory effects of both of these substance on the basal tone of ileum is probably due to reduced Ca<sup>+2</sup> stores and increase in cAMP content that are involved in the relaxation of smooth muscle.

## ISOLATION AND IDENTIFICATION OF PATHOGENIC FUNGUS ON ROSE RINGED PARAKEET (*PSITTACULA KRAMERI*) WOUNDED CHILI AND GUAVA FRUITS

### Hina Zafar and Syeda Azra Tariq

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Chili (Capsicum annuum L; Solanaceae) grow in tropical climate which is ideal for the development of opportunistic fungi while, Guava (Psidium guajava) is a popular fruit; gives two crops a year and widely cultivated in Pakistan. Rose ringed parakeet, Psittacula krameri is one of the most destructive bird pest of orchids and horticultural crops. It damages the fruits by gnawing and wounding which is exposed to many pathogens resulting in heavy losses in crop yield. After parakeet invasion the wounded chilies are wide-open to fungal attack which spread rapidly to all over the plant causing enormous damage. In this regard a study was carried out to discover the intensity of fungus development in parakeet wounded chili pods and guava fruits in comparison to undamaged pods and fruit. For the purpose 20 wounded samples of chili pods were collected randomly from 5 chili plots of agricultural fields at Gadap. Result disclosed that out of 20 samples 9.85 samples were found positive for the fungus pathogenesis. Out of these samples 4.25 were positive for Alternaria solani; 3.74 were positive for Colletotrichum capsici and 1.86 were positive for Fusarium oxysporum. No fungus growth was observed in control (Un-wounded chili pods). Furthermore, 20 samples of guava fruit were also collected randomly from guava agricultural fields at Gadap. 8.85 samples were found positive for the fungus pathogenesis. Out of these samples 3.25 were positive for Alternaria alternat; 2.42 were positive for Rhizoctonia solani; 1.32 was positive for Aspergillus flavus and 1.86 were positive for Fusarium sp. No fungus growth was observed in control (Un-wounded Guava fruit).

## BIOLOGICAL PARAMETERS OF SPODOPTERA LITURA ON DIFFERENT HOST S UNDER LABORATORY CONDITIONS

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Spodoptera litura is polyphagous insect causing immense damage to various crops including cabbage, cotton, okra, alfalfa and sesbania throughout the world. The rearing of *S. litura* was carried out under controlled laboratory conditions (25±2°C, 65±5% RH and 14:10 L:D) during 2019 to check its biological parameters on different hosts. The biological parameters such as fecundity, fertility and survival rate were highly affected by tested hosts. *S. litura* females oviposited most on cabbage and okra, least on sesbania and intermediate on alfalfa. Among hosts, the highest survival rate of larvae was recorded on cabbage (81.5%) followed by okra (70.6%), alfalfa (63.6%) and sesbania (51.4%). The larvae fed on cabbage leaves supported highest adult success i.e. 70.81 percent and lowest mean adult success was observed on sesbania (36.00%). The maximum mortality was recorded in the pupal stage (30.12, 23.47, 12.45 and 7.41%) on sesbania, alfalfa, okra and cabbage, respectively. Pupal weight was found lowest on sesbania and alfalfa while highest on okra and cabbage. The weight of male pupa was less than female pupa. The study resulted that male was long lived than female.

### RESPONSE OF HOT SPICES ESSENCES WITH METHYL EUGENOL TO ATTRACT *BACTROCERA* SPP. AT JUJUBE ORCHARD

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Fruit flies found in tropical Asia and has been spread in several tropical countries of Asia. *Bactrocera* spp. are serious threat to production and quality of fruits particularly Jujube, Chiku, Mango and Guava in relation to Sindh Province. Male adult population of fruit fly species captured through hot spices essences were tested with Methyl eugenol i.e. Cinnamon essence (T1), Veneta cucine essence (T2), Black peeper essence (T3) and control Methyl eugenol (without any essence) (T4). All were installed at Jujube orchard Agriculture Research Institute (ARI) Tandojam. Regardless the traps *Bactrocera* spp. infestation was highest at Black peeper essence with methyl eugenol pheromone traps; while traps installed at veneta cucine essence, control Methyl eugenol (without any essence) could not catch *B.zonata* and *B.dorsallis* more than Cinnamon essence.

### FEEDING OF ROSE-RINGED PARAKEET (*PSITTACULA KRAMERI*) ON SUNFLOWER IN CENTRAL PUNJAB

### Muhammad Umer Mukhtar\*, Muhammad Afzaal Akram, Hammad Ahmad Khan, Mobeen Azam, Amt-Un-Nisa and Muhammad Waqar

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This study regarding the damage patterns of sunflower caused by the rose-ringed parakeet on one-acre crop was extended for a period of three months (March through May). In the different weeks of the present study, variable values were recorded, like in March (204+1.75) and other depredatory percentages were recorded. It was evident that the damage intensity was high following the hiatus of the previous night. Due to its high significance as an oil seed crop, the parakeet problem remains important to prevent damage and obtaining the good quality of oil to satisfy the domestic oil requirements of the country.

### SEASONAL MONITORING OF *BACTROCERA* SPP. THROUGH METHYL EUGENOL TRAPS IN JUJUBE ORCHARD

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Fruit flies of genus *Bactrocera* are commonly associated to Jujube, Guava and Mango orchards. *Bactrocera* spp. are considered the most destructive insect pests of fruits and vegetables in the world. Male adult population of fruit fly species captured through methyl eugenol pheromone traps installed at different heights i-e surface, 1 metre, 2 metres and 3 metres were installed at Jujube orchard Agriculture Research Institute Tandojam. *Bactrocera* spp. infestation was highest when the methyl eugenol pheromone traps were installed at 2 metres height; while traps installed at 1 metre and 3 metres height could not catch *Bactrocera zonata* and *Bactrocera dorsalis* more than surface installed traps.

### FORAGING RHYTHMS OF SOME BIRDS IN AN AGRO-ECOSYSTEM OF FAISALABAD

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Studies regarding the foraging rhythms of some birds in an agro-ecosystem of Faisalabad comprising the parakeet, sparrow, myna and crows were sampled on the weekly intervals from February through May. A total of 16-weeks was spent to make such observations. Seemingly, the house sparrow appeared to be the highest depredating bird  $(92\pm1.85)$  followed by the rose ringed parakeet  $(61\pm1.47)$ , crow  $(50\pm1.28)$  and common myna  $(37\pm1.15)$ . During all these weekly observations, the movement patterns which were recorded foe all such birds differed to some extent, perhaps depending on their feeding opportunities and their regular movement patterns from and to the food resources from their roosts. Conclusively, it was suggested that knowing their movement patterns in detail, possibly some management measures (environmentally sustainable) can be applied rather than incorporation of use of chemicals to reduce their depredations.

### SEASONAL DISTRIBUTION OF DINOFLAGELLATE (NOCTILUCA SCINTILLANS) IN THE NORTHERN ARABIAN SEA

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The present study reports seasonal distribution of dinoflagellates from Northern Arabian Sea (Sandspit). Sea water was collected using Niskin water sampler from two stations for water quality and nutrients analysis. *Noctiluca scintillans* (dinoflagellate) is cosmopolitan in distribution and occurs in coastal environment. It is omnivorous, symbiotic and feed on planktons, eggs of fish and detritus. Maximum number of *Noctiluca* was observed 25240 cell/L in August than 2400 cell/L in December and 680 cell/L in January. Among toxic species of dinoflagellates, *Ceratium fusus, Dinophysis caudata, Gyrodinium spirale, Prorocentrum micans, Prorocentrum arcuratum* and *Gonyaulax spinifera* were observed. The dinoflagellates are always present in water and produce bloom when population explosion occurs due to salinity, temperature, excessive nutrients and anthropogenic activities. Some organisms produce toxins that affect filter feeding animals such as clams, oysters, mussels and crustaceans. Red tide caused fish kill, affect fishery resources, biodiversity and threat to the marine environment. The increased frequency of algal blooms in the coastal areas worldwide is mainly due to human activities. As algal blooms cause hazardous impacts on coastal areas, economies and public health, so Government should develop strategies for controlling pollution.

# FIRST CONFIRMED RECORDS OF *DUPLICARIA SPECTABILIS* (HINDS, 1844) (NEOGASTROPODA: TEREBRIDAE) FROM BALOCHISTAN, PAKISTAN

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Mollusca are diverse group of the animal kingdom, widely distributed throughout the world. The largest class Gastropoda accounts for 80% of the total known living species. The neogastropod family Terebridae Bruguière, 1789 are distributed in tropical and subtropical waters worldwide. It is a large family of about 400 valid species recorded thus far and some twenty species have been reported from Pakistan. Members of this family are infaunal, inhabiting

sandy, muddy and/or silty substrate, from the intertidal zone to a depth of 1,000 m. Several previous studies of Sonmiani Bay lagoon did not mention terebrid species. Only *Duplicaria duplicata* (Linnaeus, 1758) have been reported from the West Bay of Gawadar. In the present study we report on the first records of *Duplicaria spectabilis* (Hinds, 1844) collected from sandy/muddy lagoon at low tidal zone during surveys at Sonmiani Bay (Miani Hor) and Ormara (Taq Bay) along Balochistan coast. Besides reporting the new record from these areas with relevant information on species description will aid identifications and studies of the malacofauna of Pakistan.

# EVALUATION OF ANTI-CARCINOGENIC POTENTIAL OF HYDRASTIS CANADENSIS (GOLDENSEAL) AGAINST CERVICAL CANCER

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Cancer is the second leading cause of death after heart diseases and cervical cancer is the most common gynecological cancer after uterine carcinoma in developing countries. Hydrastis canadensis is extensively used in complementary and alternative system of medicine to treat various ailments. The study was aimed to evaluate the anti-carcinogenic potential of Hydrastis canadensis against cervical cancer. Cytotoxic activity of ethanolic extract of plant against HeLa cells was assessed by using MTT assay. Different doses of plant extract ranging from  $2.5\mu g/ml$  to  $100\mu g/ml$  were applied to check the IC50 value. The IC50 of plant extract on HeLa cells was  $23.4\mu g/ml$ . The results indicate that Hydrastis canadensis extract has cytotoxic effect on HeLa cells and supports its use as an alternative of chemotherapy to treat cervical cancer.

#### EFFECTS OF FERTILIZER ON INSECT POLLINATORS AND CROP YIELD

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Pollination is essential for the seed and fruit setting and produce of the crop even in self-compatible crops. Plant nutrient directly or indirectly affect pollination and its pattern, pollinators attraction and behavior and crop. Plant fertilizers have ability to influence the plant and pollinators relationship which ultimately affect the crop yield. Yield is highly dependent on plant nutrition and soil fertilization. The response of plant or crop to the fertilizer is species specific. Day-by-day increasing application of the fertilizer for increasing the crop produce not only polluting the soil but also decreasing the visitation rate of the pollinators. Among the pollinators, bees are the most adversely effected species due to non-judicious use of fertilizers. Studies found that plants with high application of fertilizer not only decrease the pollination but also cause mortality in them because of the changing in chemical composition of the plants. High dosage of Nitrogen (N) fertilizer decrease abundance of the pollination while 25% yield increased at moderate level of N in sunflower. Increase in the vermicompost rate decrease pollinator's visitation time in cucumber and have no effect on yield. Low level of fertilizer showed positive effects on common beans yield and its pollinator, *Apis melifera*. In a grassland plant species high level of fertilizer decrease the level of amino acid and cause negative effects on bumble bees, *Bombus terrestris*. Hence it is concluded that by the judicious use of fertilizers we not only conserve the native pollinators but also enhance the pollinators services which ultimately increase the yield.

#### **DISEASES IN SOLITARY BEES**

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Pollination is essential for crops production as 2/3 of crops reproduction depends upon the pollinators. Worldwide value of insect pollination is \$175 billion per year. There are many different types of insect pollinators such as bees, flies, beetles, moths, and butterflies. In different flowering plants, solitary bees are common flower

visitors. Small number of ecosystems, habitat deterioration and sub-lethal pesticide is the reason decline the bee's population. Chronic, dietary and neonicotinoid exposure has severe harmful effects on solitary bee reproductive output. According to a survey, due to pesticide daily use, forager mortality rate is 35.5% in stable colony. Neonicotinoid negatively affects the memory and learning abilities, for example homing and foraging behaviors. Losses due to disease are one of the primary reasons of decline in the biodiversity of pollinating bees' population. Several types of diseases and pathogens attack on solitary bees, honey bees and wild bees. These diseases are fungal, bacterial and viral. Chalk brood disease attack on honey bee colony and destroy whole colony worldwide. In managed bee populations, fast-evolving RNA viruses, cause severe colony losses and also threaten economically and ecologically, also in wild pollinator communities. Viral infections have been raised as a potential cause of colony failure syndrome. This is difficult to estimate the losses of wild bees due to diseases because of negligence and lack of research and interest. Therefore, it is concluded that management policies, trainings and other practices should be considered that overcome the chances of these diseases. So, it is the future need that we should minimize habitat loss, attacks of parasites and pathogens to conserve the wild bees for stable ecosystem in upcoming days.

# PREVALENCE AND DISTRIBUTION OF HUMAN *PLASMODIUM* INFECTION IN DISTRICT SANGHAR, SINDH, PAKISTAN

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Malaria is the most common mosquito borne disease, which is transmitted to the human through the bite of the female anopheles mosquito carrying the parasite (Plasmodium). *Plasmodium vivax* and *Plasmodium falciparum* are prevalent in Pakistan. In the present study data of malaria and blood samples of suspected patients were collected for detecting malaria parasite species during January 2018 to December 2018, from different hospitals of six localities of district Sanghar. Total 17154 suspected patients were observed, out of these 315 were found malaria positive. Among these positive cases of malaria 291 were identified *P. vivax* while 12 were *P. falciparum*. In district Sanghar highest ratio of parasite species was founded in *Plasmodium vivax*.

### FIRST RECORD ON THE SPECIES OF GENUS *AGRIOTES* ESCHSCHOLTZ (1829) (COLEOPTERA: ELATERIDAE: ELATERINAE) FROM KHAIRPUR SINDH, PAKISTAN

#### Shabana Mangi, Waheed Ali Panhwar, Abdul Manan Shaikh, Hafeeza Gul and Muhsan Raza

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Click beetles belong to order Coleoptera. Click beetles are usually cosmopolitan beetles and recognized by specialized process of click mechanism. They are simply identified by their lengthened body, round figure, generously distinctly prothorax, posterior angles of pronotum are rounded, a prosternum covered with spines, click on the mesosternum producing aggressive voice for defense clicking produced, antennae eleven segments, lengthened pointed. This family can be distinguished for the adult's capability to click. The adult's click beetles feed upon flowers and grasses, below the woof, within decomposing timber and mostly feeding underground roots. Larvae of click beetles are usually saprophagous, living on dead material. Extensive surveys were carried out to collect Click beetles fauna of Sindh province. *Agriotes sameki (Platia 2003) was* belong to subfamily Elaterinae, Elaterinae is the largest subfamily and monophyletic group, it contain 9 tribes, 180 genera, *Agriotes sameki* (Platia 2003) identified on the taxonomical characteristics including body coloration dark brown to reddish brownish coloration, head clypeus, para clypeus deeply brown, antennae reddish brown, pronotum, humeral angles dark brown, humeral angles tips black, Scutellum, abdomen deeply brown, elytra black, ventral side light brown, head (0.4mm) triangular para clypeus longer than clypeus convex in shape, clypeus concave, pronotum(10mm) convex, posterior margins concave, dorsal surface convex, antennae 12 sections, 2<sup>nd</sup> section thick longer than other section, cyclinderical in shape, 3<sup>rd</sup> smaller than 4<sup>th</sup> section, 5<sup>th</sup> section longer than 6<sup>th</sup> section, middle sections triangular in

shape, last one section small and triangular in shape, humeral angles pointed, tips concave, space between humeral angles and abdomen, densely punctuations on the complete body with different locations and measurements, Scutellum small rounded shield theca like structure, abdomen (u) shape structure, elytra(1.4mm)anterior side convex, posterior side concave and denticated, dorsal surface convex with lateral lines with punctuations, lateral lines with different measurements with various location, ventral side light brown, legs (femora) tarsi cyclinderical, tibia denticated or teeth like structure., total body length(1.9mm).

### STUDY ON TAXONOMY OF OWLET MOTHS (NOCTUIDAE: LEPIDOPTERA) FROM DISTRICT KHAIRPUR MIRS

#### Paras Soomro, Abdul Manan Shaikh , Waheed Ali Panhwar and Fateh Muhammad Soomro

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Moths belongs to order Lepidoptera are phytophagous insects and are considered as prime insects in the world and are distinctive group of insects in ecosystem, consuming plants, larvae as well as adult feed on nectar. Noctuidae or owlet moths are group of robust moths. The name of this family is taken from the way that night species have eyes, with reflect light with and an orange gleam. Most species are dim to cocoa in color frequently with dim and light spots on the forewings. They are nocturnal and are normally attracted to lights those larvae of Noctuidae have tufts of short abounds and might be brilliantly colored. Most are foliage feeders and the insatiable hungers of few species have turned them to serious pest of wide range of crops and thus they are known as army-worms and cutworms. Extensive surveys were carried out to collect Noctuidae fauna of distrct Khairpur Mirs. A total of 221 specimens were captured and were identified into 11 genera and 15 species of Noctuidae i-e: Mythimna unipuncta, (Haworth, 1809), Mythimna impure (Hübner, 1808), Heliothis adaucta Butler, 1878, Heliothis peltigera (Denis & Schiffermüller, 1775), Agrotis ipsilon (Hufnagel, 1766), Agrotis exclamationis (Linnaeus, 1758), Meterana pansicolor (Howes, 1912), Meterana alcyone (Hudson, 1898), Spodoptera litura (Fabricius, 1775), Grammodes stolida (Fabricius, 1775), Graphania pagaia (Hudson, 1909), Helicoverpa armigera (Hübner, 1808), Heliothis armigera (Hübner, 1808) Dipaustica epiastra (Meyrick, 1911) and Dysgonia algira, Linnaeus in 1767. Beside this, description for species along with digital images were also provided for easily identification of species. Present study is an initiative step towards the biodiversity of Noctuidae fauna of district Khairpur Mir's.

### TAXONOMY AND DISTRIBUTION OF GENUS *GRYLLOTALPA* LATREILLE, 1802 (GRYLLOTALPIDAE:ORTHOPTERA) FROM NARA DESERT KHAIRPUR SINDH

#### Sameet Kumar and Waheed Ali Panhwar

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Nara desert is the one of the biggest desert of Pakistan. It is located in Taluka Nara and District Khairpur Sindh Pakistan. It has a dry weather and beautiful scenery; it is very appealing to nature lovers. The verity of habitat of Nara Desert gives it a special climate, with winter temperature below zero, summer temperature above 50°C and the climatic conditions of this area make it more suitable for the biodiversity of insects. The mole crickets are members of family Gryllotalpidae Order Orthoptera. The cricket species are omnivorous. The mole cricket lives underground, making burrows and feeding on plant roots, larvae and other insects. It goes to the surface only at night-mostly in the mating season. It can fly too, when changing territory or when females are searching for males. Males call females by chirping. They are considered a pest in some regions. During present study extensive surveys were made to collect the mole crickets belonging to genus Gryllotalpa and about 232 specimens were captured falling into five species i-e: *Gryllotalpa africana*, *Gryllotalpa hirusta*, *Gryllotalpa minuta*, *Gryllotalpa orientalis* and *Gryllotalpa ornata*. Additionally, taxonomic characters were noted down. A key for distinguishing the mole crickets in Nara desert Khairpur and a

distribution map for this region are presented. Finding of this study will definitely helpful for the scientists dealing with this group.

# ON THE IDENTITY OF *PHEIDOLE NEITNERI*, EMERY 1901 FROM TALUKA GAMBAT, SINDH PAKISTAN

#### Shahnawaz Ujjan, Waheed Ali Panhwar, Javed Ahmed Ujan, Khadim Hussain Memon and Zaib-un-Nisa Memon

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The ants are most diverse group of insects falling in order Hymenoptera and family Formicidae. They are regarded as an important insects in building ecosystem. During the extensive surveys 121 specimens were captured by keeping traps the samples were killed by standard entomological procedures and then preserved into insect boxes. The material was identified into single species i.e: *Pheidole neitneri*, Emery 1901. Important morphological features were observed under stereoscopic binocular microscope. The material was then preserved for further studies.

# SOME NEW RECORD OF SCORPION FAUNA FOUND IN DIFFERENT REGIONS OF DISTRICT PISHIN (BALOCHISTAN) PAKISTAN

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Study on diversity of scorpion was carried out from April to October, 2018 in district Pishin, (Balochistan), Pakistan. Specimens were caught in field at night using torch light, and under stones, cracks of mud wall in day time. A sum of 130 (79 males and 51 females) scorpion species were collected and identified based on main diagnostic features. Five species from the family (Buthidae) are recorded from this region of Pakistan for the first time. The identified specimens include Andructonous rubustus, A. austrialis Mesobuthous cypirus, M. eupeus, Hottentotta (Buthotus), Buthacus (Birula), and Tityus serrulatus. The Androctonus austrialis showed high ratio of abundance 41(70.73% male, 29.26% female) followed by Hottentotta (Buthotus) sp. 31 (61.29% male, 38.71% female) and Buthacus (Birula) sp. 24 (62.5% male, 37.5% female) respectively. Among six localities, Pishin town showed high number (29) of scorpion species followed by Khanozai (26) and Baostan (25). Based on male specimen's measurements, Hottentotta (Buthotus) sp. was found to be the larger scorpion with 11.1 cm total body length compared to the male specimens of the remaining four species. Color comparison indicated four species (A. austrialis, A. rubustus, Buthacus (Birula) sp., Tityus serrulatus) with yellow to yellowish body color, whereas M. cyprius, M. eupeus and Hottentotta (Buthotus) sp. showed yellowish brown, gray, and black to yellow body color respectively. Findings concluded that foremost and diverse assortment of scorpions in the study region are existed. Since arachnids exhibit restricted movements and are vulnerable to habitat modification.

### LENGTH-WEIGHT RELATIONSHIP AND CONDITION FACTOR FOR FISH SPECIES FOUND IN LOWER ZHOB RIVER, NORTHERN BALOCHISTAN, PAKISTAN

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This research study was conducted with the aim to evaluate the length-weight relationship (LWR) parameters and condition factor (K) for three native fish species, namely: *Labeo rohita* (Hamilton, 1822), *Cyprinion watsoni* (Day, 1872) and *Garra gotyla* (Gray, 1830). A total of 306 specimens including 251 *L. rohita*, 255 *Cyprinion* 

watsoni. and 250 G. gotyla were caught by scoop net in the months of February, March, and September, October 2019 from the lower Zhob River, in northern Balochistan. The estimated total length and total weight for these specimens were ranged from 7 to 18.9cm and 5 to 79g; 6.8 to 18.9cm and 2 to 73g; 7.5 to 17.6cm and 5 to 68g respectively. Based on the results, the values of b varied between 1.25 for  $Garra\ gotyla$ , 1.34 for , to 3.19 for  $Cyprinion\ watsoni$ . All length-weight relationships were significant (P<0.001), with r greater than 0.97. The condition factor (K value) were found to be between  $0.035\pm0.034$  for G. gotyla to  $0.20\pm0.081$  for L. rohita to  $0.33\pm0.11$  for C. watsoni. These are the first estimated parameters of length-weight relationships for the fish species live in the Zhob River basin.

# STUDY ON THE PATHOLOGICAL ORGANS AFFECTED WITH BACTERIAL DISEASE (SALMONELLA PULLORIUM) IN BROILER BIRDS OF HYDERABAD DISTRICT

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Chicken meat is a main supply of high-quality proteins, minerals; vitamins are containing the benefit onto the red meat. The current poor nutritional status in the country is due to lack of sufficient availability of animal protein in the food. Poultry is the cheapest available source of animal protein with high biological value for our masses. The diagnosis of the poultry diseases as well as in the immediate analysis of the feed and finding of the mycotoxins in the feed are important. Various poultry diseases are more successfully or efficiently controlled by vaccination. In Pakistan, higher mortality in poultry is attributed to poor management unsuccessful health treatment programs and sever outbreak of different bacterial and viral diseases. In Pakistan, these diseases are commonly occurring throughout the year causing heavy economic losses. Clinical diagnosis of the disease before the occurrence of mortality is difficult, since the birds do not show any specific clinical signs. Diagnosis has been carried out on the basis of gross and histopathological changes in lesions. During postmortem examination affected birds were examined for pathological lesions from different organs i.e. Liver, spleen, heart, lungs with enlargement, discolouration, necrosis, hemorrhages was found infected with Salmonella Pullorum with different visceral lesions i.e The variable rate of occurrences of pathological lesions in different visceral organs with pullorum disease was also observed at different commercial broiler farms of yderabad district.

### GENE THERAPY, ELECTROPORATION, AND THE FUTURE OF WOUND-HEALING THERAPIES

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Long-term injuries are fatal major medical accidents. A cure usually helps and incurs adequate health costs. It is foreseeable that 1.25% of the U.S. population is burned each year, while 6.5% of the population suffers from long-term skin ulcers caused by diabetes, stress and venous stasis. The recovery of Gush's disease is a long and complex process that usually takes a year to fully cure. It is estimated that the cost of treating poor foot wound healing in the United States is \$ 1 billion per year. There are many current commercial products on the market to provide a favorable environment for problematic open wounds. Local platelet-derived growth factor (PDGF) -BB has been shown to be effective in improving the healing of damaged wounds, but it has the disadvantage of requiring large and repeated doses. Recently, researchers have focused on the possibility of inserting genes encoding growth factors such as PDGF-BB into cells involved in wound healing responses. This approach offers the potential of a single dose of growth factor to treat chronic wounds. There are several methods for gene insertion, including viral vectors, gene

guns and electroporation. This article focuses on electroporation and reviews the strategies and potential of these methods.

# PREVALENCE OF HUMAN MALARIAL PARASITES IN TALUKA ROHRI, DISTRICT SUKKUR

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The study was designed to investigate the *Plasmodium* prevalence in human population of taluka Rohri, from January to December 2018. The analyzed data was based on the month, age and sex wise. Out of 1230 observed cases of malaria 9% were reported positive. *Plasmodium vivax* 82.5% and *Plasmodium falciparum* 17.5% were reported out of positive cases. *Plasmodium vivax* was described 75% in April, October and November, and 35% in January and December. While, *P. falciparum* was recorded as the highest 65% in January and December and it was not recorded in the April, October and November. Prevalence of *P. falciparum* in male and female was recorded lower, than prevalence of *P. vivax*. The *P. vivax* prevalence was highest 75% in 11-20 year age category and the lowest 70% in 1-10 year age category. However, *P. falciparum* infection was found highest 27% in 1-10 year age category and lowest 25% in 11-20 year age category.

### A REVIEW ON CURRENT STATUS AND CONSERVATION STRATEGIES OF MIGRATORY BIRDS IN PAKISTAN

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The bird fauna of Siberia travels to the eastern region through different flight areas to avoid the danger of extreme cold winters every year. The Indus Flight Area of Pakistan is a Central Asian flight route for migratory birds related to Central and East Africa flight routes. Of the billions of people who spend the non-breeding season in the tropics, there are more than 500 species of birds. These birds live in almost all habitats, even those that have been severely altered by humans, such as agricultural ecosystems, and play a vital role in tropical ecosystems. The decreasing trend of migratory birds recorded from Pakistan's freshwater reservoirs is mainly attributed to the loss of wintering habitat, habitat change, fragmentation, eutrophication and illegal hunting by local and international hunters. Some other factors documented are pesticides, eutrophication, pesticide pollution, heavy metal pollution, wetland pollution, and invasive species. There are multiple reasons for this adverse effect, due to the large number of birds visited during the winter, including tail-tailed eagles, black storks and water Pi, seagulls, hawks, terns, ducks and Cor, painted storks and Dalmatian pelican Black-tailed gods and fish and birds, egrets practice ducks, little egrets and night herons, wild geese, ducks, siberian cranes and Bu, etc., lead to a decline in species richness. The protection of migratory birds has become a major focus of global bird protection efforts, the main purpose of which is to protect their populations rather than prevent extinction, but this is very difficult, as habitat loss and degradation during breeding, wintering, and migration halts habitats can cause population cut back. Therefore, the protection of migratory birds requires large-scale conservation work that takes into account the migration links between breeding and winter farms, the possibility of effective protection, at least in some agricultural ecosystems, and the possibility of obtaining pesticides and pathogens across borders Sex. Regular surveys of important migratory birds to assess their migration status, quantity and migration methods are another important step in protecting birds. In order to effectively protect migratory birds, integrating various methods to protect their habitats, preventing illegal hunting and poaching, reducing agricultural and industrial pollution, and preventing heavy metal poisoning in reservoirs are important considerations. The cessation of human activities and popular awareness programmes in electronic and print media may be effective protection strategies during their stay in Pakistan.

# ASSESSMENT OF VARIOUS PLANTS EXTRACTS TO CONTROL THE SUCKING PEST JASSID (AMRASCA SPP) ON BRINJAL (SOLANUM MELONGENA) CROP

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In this experiment the main purpose is to screen out the bio-pesticides which is safe for human health and environment. In this experiment different biopesticde were used to control the jassid. The data were recorded after the 24 hours, 48 hours, 72 hrs and 7 days of treatment application. The RCBD (Randomized complete block design) with three replicates were used. The extracts of different biopesticde were used with the 18 % solution. The different treatments, Neem seeds, Alovera extract, Moringa leaves Extracts, Ginger Extracts, Lemon seed extracts, Eucalyptus. The minimum populations were recorded 0.98/leaf against Neem Extracts and highest populations were recorded on the Moringa leaves extracts which was 4.5/leaf. The biopesticides Eucalyptus and Ginger showed good results. The result showed that Neem extract is better with minimum post treatment infestation for field application. The means were compared by Duncan's Multiple Range Test (DMRT) at P = 0.05. IBM.

# FIRST RECORDS AND MORPHOLOGICAL IDENTIFICATION OF THE SEAHORSE *HIPPOCAMPUS KELLOGGI* JORDAN AND SNYDER, (1902) IN COASTAL WATERS OF PAKISTAN

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The first records of seahorse *Hippocampus kelloggi* from the coastal waters of Pakistan based on morphological and morphometric characteristics. The seahorses representatives of the family Syngnathidae, which that's includes pipe fishes, seahorses, pipe horses and sea dragons. There are about 50 species reported across the world and they have been found to inhabit coral reefs, seagrass beds and also coastal mangroves. All seahorse species (*Hippocampus sp.*) are included in the IUCN Red List (http://www.iucnredlist.org). *H. kelloggi* is also listed as data deficient since 2003 by IUCN, Red List Vulnerable (VU) (2012) and now the global conservation status of *Hippocampus kelloggi* is classified as IUCN Red List Category & Criteria: as a vulnerable A2cd ver. 3.1 (2017). Previous record of *Hippocampus kelloggi* of the Indo Pacific region, including China; India; Indonesia; Japan; Malaysia; Philippines; Thailand; United Republic of Tanzania; Viet Nam. The current study showed the presence of *H. kelloggi* from the coastal waters of Pakistan and faces similar threats as other Hippocampus species. The protein pattern of species was also determined through SDS page electrophoresis.

# A NEW RECORD OF SEA FAN *ECHINOMURICEA INDICA* (ANTHOZOA; OCTOCORALLIA) FROM THE PAKISTAN COAST

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Corals are the sessile invertebrate animals belongs to the phylum Cnidaria. Sea fans, a marine coral belong to subclass Octocorallia (eight-branched tentacles and eight septa in their polyp structure) and order Alcyonacea (soft corals). The Sea fans are colonial sessile animal having tough colonies which are flexible along the central axial rod that covered with a living surface called a rind. The axial skeleton of the colony is composed of a fibrous protein substance, known as gorgonion. Thus, the common names, Gorgonian, Horn Coral or Horny Coral, are derived. They

capture planktonic organisms and microscopic food particles from the water column and can absorb dissolved organic matter. They are widely distributed in the and abundant in the shallow waters' areas of the Indo-Pacific regions of the world. Along Pakistan a true coral reef ecosystem is not found instead a patch of reefs are located mainly along the Baluchistan coast. During the present study, a new record, *Echinomuricea indica* Thomson and Simpson, 1909 from Pakistan. The morphological characters of the species that were analyzed included the colony shape, branch shape and the size and composition of the sclerites. Collected colony was ovate or semicircular in shape. Branches of colony arise directly from the stalk; identity loses after a short distance and the branchlets formed often transverse in a radial pattern. Sclerites are varied in shape that includes thorn star sclerite and scaphoid type. This new record contributes to understanding the diversity and distribution of the octo-corals along the North Arabian coast of Pakistan.

### CRANIOMETRICAL ANALYSIS OF ASIATIC GOLDEN JACKAL FROM SOUTHERN PUNJAB, PAKISTAN

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The current study reports the craniometric analysis of the golden jackal (*Canis aureus*) from Dera Ghazi Khan, Punjab, Pakistan. The study reports the 24 linear skull and dental parameters and six craniometrical indices of golden Jackal. The analysis included 10 individuals (8 males and 2 females), originating from 3 sympatric populations with variable density. Age variation, sexual dimorphism and cranial variation were assessed in order to resolve the phenotypic similarity of the *Asiatic* golden jackal populations. The skulls of jackal were collected as dead animals from roadside accidents. The skulls were cleaned by entombing in the ground. After two months ground was dug and skull were recovered. Furthermore, the skulls were cleaned with NaOCl to remove the smell and tissues. A total of 60 cranial parameters were taken into consideration for assessment of the intra-specific variations among the golden Jackal population. All the data were analysed with the help of SPSS software, students T-test and ANOVA. The intra specific variations were calculated (p>0.05). The insignificant differences represent no intra-specific variation and favored the inclusion of the species *Canis aureus*. The study reports first craniometrical analysis of golden Jackal from Pakistan and add significant data in the available data set of Family Canidae.

### USING DIFFERENT TYPES OF TRAPS FOR MONITORING ARTHROPODS DIVERSITY IN SUB-TROPICAL REGION OF PAKISTAN

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Installation of traps in agricultural field is economically most important and cheaper techniques to observe arthropod's diversity. Nowadays, cost-effective ecological monitoring of arthropods by traps has been gaining interest in the field of environmental entomology for the last few decades. This study explains the effectiveness of four different types of traps (pitfall, yellow-sticky, pan, and PVC barrier traps) to monitor the arthropod diversity in summer and winter seasons. These traps were installed in different mango orchards located in Punjab Pakistan. The trap type and collection season had significant impact on the abundance and diversity of arthropod groups/orders.

Arthropods diversity was captured 1.5 times higher in summer than in winter season. However, among traps, the pitfall traps were most effective than other traps for arthropod assemblage in both seasons. The pan traps were found most effective in summer season, while sticky traps in winter season. The pitfall traps exhibited highest taxa richness index values (8.00 for summer and 5.00 for winter seasons), while the lowest values were recorded for barrier traps (5.00 for summer and 3.00 for winter season). The pitfall trap has appeared to be the most effective traps for capture or collection of Arachnida, Coleoptera, Hymenoptera, Lepidoptera, Orthoptera and other arthropods. The PVC barrier and sticky traps most effective for Dipteran and Hemipteran's insects, respectively, and hence, are recommended for ecological monitoring of these arthropod groups in future studies.

### TAXONOMIC STUDY OF ZYGOPTERA (ODONATA) FROM DISTRICT MULTAN, PUNJAB, PAKISTAN

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Zygopteran species are predatory in nature play an important role for the development of biological control, both the nymph and adults of the zygopteran feed on small insect like jassid, white fly, thrips, psyllid and eggs of lepidoteran. Taxonomic study of insects plays a key role in proper identification and classification of insects. In the present study damselflies ere collected by comprehensive surveys conducted in different localities of district Multan during March to May 2019. Collected specimens were killed in Potassium cyanide and pinning, tagging and preserve properly. For the extraction of genitalia, abdomens of the specimens were detached and dip in 10% KOH solution for 24 hours. Dissection of abdomens was made under stereo microscope. Genitalia were dip in different concentration of ethanol like 50%, 60%, 70% respectively for more clearance. As the result of total five species were identified on the base of genitalia and morphology. These five species belong to four genera of family coenagrionidae. Two species like, *Ceriagrion coromendelianum* and *ceriagrion olivaceum aurantiacum* belong to genus Ceriagrion, one species i.e. *Agriocnemis pygmaea* belong to genus Agriocnemis, one species like. *Pseudagrion pilidorsum pilidorsum* belong to genus Pseudagrion and one species *enallagma aspersum* belong to a genus Enallagma. Photography of adults and genitalia was also done.

# EXTRACTION OF DNA FROM FINS OF FISH BY PHENOL/CHLOROFORM/ ISOAMYLE ALCOHOL METHOD

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DNA isolation is one of the fundamental techniques that are routinely practiced in molecular biology laboratories. Till date, numerous methods have been developed for genomic DNA extraction from tissues of various types. The aim of this study is to identify the most rational technique for the isolation of high quantity and quality of DNA from fish fins. A total of 15 fish individuals were sampled for DNA extraction. After collecting the fish, it has anesthetized to isolate fin tissues. The fins were kept in polythene bags and stored in ice box for transportation to the laboratory. Then the DNA was extracted by Phenol/Chloroform/Isoamyle Alcohol method. The isolated DNA was run in agarose gel electrophoresis to assess the quality. The gel was visualized in the gel documentation chamber. The quantitative and qualitative assessment of the DNA samples was also done by using NanoDrop 2000

Spectrophotometer. Extracted DNA concentrations range from 21.7 to 7.40  $ng/\mu L$ . The result showed that the Chloroform/Isoamyle method is very effective method to achieve quality DNA with medium budget.

#### SEROPREVALENCE OF RUBELLA VIRUS IN PREGNANT WOMEN IN DISTRICT KARAK

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An observational study was carried out in district Karak to investigate the prevalence and risk factors associated with RUBV infection among pregnant women. Sample was collected by attending different health centers of district Karak. Total of 200 samples were collected and all samples were analyzed for RUBV antibodies. At overall 16% pregnant women were positive for rubella IgG antibodies. Along the age prevalence of rubella infection increases among pregnant women. Analysis of RUBV infection among pregnant women based on their education showed that prevalence of RUBV was very high in Illiterate (21.05%) as compared to graduated (8.88%) women. Housewives were more proven to rubella infection as compare to farming and employed women. Similarly women of ruler area (19.7%) showed high positivity than urban area (7.93%) for rubella virus infection. The RUBV has a very high prevalence and taken an endemic form in Pakistan, so routine screening of RUBV among pregnant women is recommended.

# EVALUATION OF T CELLS COUNT IN PIV 3 INFECTED CHILDREN SUFFERING FROM COMPLICATIONS OF PNEUMONIA AND BRONCHIOLITIS

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Parainfluenza viruses are major causative agents of respiratory infections especially of lower respiratory tract infection. Young children are more susceptible to these viral infections such as pneumonia and bronchiolitis because of their naïve immune system. But in hospitals and on clinical sides there are no current tools for diagnosis of these infections. Therefore, we hypothesized that T cells count can be used as a predictive tool for diagnosis of respiratory viruses and associated complications. So the current study was designed to find out the correlation of RNA viruses with respiratory infections as well as with T cell subsets level in children with respiratory tract infections. Blood samples (n=20) were collected from those children who were diagnosed positive for PIV 3 genotypes. Whereas 20 samples were collected from normal healthy children as control group. Flow cytometry were used for analysis of blood samples to find out the T cells subsets count. The peripheral blood T cells count showed variation in PIV 3 infected children. Lymphocytes count were significantly reduced (p<0.05). The count of CD4+ T cells did not changed significantly, but the number of CD8+ T cells (p<0.05) were reduced as compared to control group. Similarly CD3+ and CD19 cells ratio were also reduced. Reduced number of CD8+ T cells were recorded in those patients that were suffered from severe pneumonia.CD4+/CD8+ ratio were also lowered (p<0.05). These studies suggested that there must be some sort of correlation among these viral agents and T cells subpopulation. So the analysis of T cells in association with viral agents and with their complications will have a great impact in the medical field.

# STUDY ON VIROLOGICAL CAUSES OF BAD OBSTETRIC HISTORY AMONG WOMEN OF PESHAWAR, KHYBERPAKHTUNKHWA PAKISTAN

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Maternal infections caused majorly by ToRCH pathogens (Toxoplasma gondii, Rubella virus, Cytomegalovirus and Herpes simplex virus) have been considered as one of the significant factors in the causation of Bad Obstetric History (BOH), which imply previous unfavorable obstetric outcomes include two or more consecutive miscarriages, intrauterine growth retardation, stillbirth, early neonatal death, and congenital anomalies. These pathogens interfere with normal process of organogenesis resulting in poor maternal and perinatal outcomes and the loss of precious lives. A total of 103 women with BOH (BOH group) and 100 normal women (control group) of age range 15-45 years were included in this study. Venous blood was collected from all the subjects and placental tissue and cord blood were collected from BOH group. Venous blood was analyzed for ToRCH IgM/IgG by ELISA and cord blood and placental tissues were processed through PCR for nucleic acid detection. Demographic and clinical data was collected through a prescribed data form. Prior consent (verbal/written) was taken from each study participant. Seroprevalence of ToRCH infections was significantly higher (58.25%) in BOH group than control group. In BOH group, IgM seropositivity for T.gondii was 11.65%, Rubella 7.76%, CMV 19.41% and HSV 31.06%, while IgG for T.gondii 17.47%, Rubella 43.68%, CMV 41.74% and HSV 54.36%. In control group, IgM seropositivity for T. gondii 2%, Rubella 1% CMV 0% and HSV 2% while IgG for T. gondii 2%, Rubella 1%, CMV 2% and HSV 1%. The PCR results of cord blood samples showed T.gondii 9%, CMV 6%, Rubella 11% and HSV 25%. 30 placental tissues obtained from aborted women of BOH group, only 15% cases (13/30 HSV, 2/30 Rubella) were positive by PCR. ToRCH frequency was higher in women with stillbirth 94%, preterm labour 88%, and lower 63% for congenital anomalies was recorded. This study shows a very strong association of ToRCH agents as a potential cause of BOH. Hence screening and early diagnosis for TORCH Infections in women with BOH can help in proper management of these cases and prevent deadly outcomes and pregnancy wastage.

#### PRODUCTION OF BIOGAS FROM SILKWORM EXCRETED WASTE

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In view of the increasing demand for organic agriculture, waste utilization and environmental protection, sericulture not only focuses on the production of alfalfa, but also on other ways that are conducive to cocoon production. The economy of the farm not only to pass, but also to find new sources of income for small-scale farmers Sales are also subject to multiple uses of by-products. Insect farming technology provides cheap Biomass sources may be good materials in biogas production. Studies have shown that the tested substrates, including silkworm breeding waste and caterpillar excreta, Produce biogas production comparable to other matrices of agricultural origin, such as cattle, pigs and pigs, Chicken manure

### FEEDING PREFERENCE AND POPULATION DEVELOPMENT OF TRIBOLIUM CASTANEUM ON DIFFERENT CEREALS UNDER CHOICE CONDITIONS

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*Tribolium castaneum* (Coleoptera: Tenebrionidae) is one of the most widespread and destructive insect pest of stored grains that cause heavy loss to the stored grains. Therefore, this study was carried out to determine the relative

preference of *T. castaneum* for different cereals under choice conditions and to assess the losses to grains because of its feeding. Five different cereals i.e., barley, wheat, maize, millet and rice along with a control (an empty box) were randomly filled in boxes in equal quantities (100 grams each). All boxes were attached together and fifty adult *T. castaneum* were released in the center of boxes to determine their preference for various cereals. Subsequent observations on the population of beetles was recorded on fortnightly basis for three months and weight loss was assessed at the end of experiment. Among treated grains, overall the highest population of adults was recorded in barley (47.17 beetles), followed by millet (43.20 beetles), whereas, maize (2.17 beetles) suffered the lowest population development of *T. castaneum*. Considering the population of *T. castaneum* in various cereals, the highest and lowest weight loss was recorded in barley and maize. respectively. Keeping in view of differential preference of *T. castaneum* on different cereals, it is suggested that proper arrangement should be done to store cereals separately to restrict the movement and losses of *T. castaneum*.

#### CRANIOMETRICAL ANALYSIS OF EQUUS (EQUIDAE: MAMMALIA) FROM PUNJAB, PAKISTAN

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The current study has been carried out on 15 adult (males and females) skulls of family Equidae taken from zoological museum, University of the Punjab, Lahore. The purpose of this study was an estimation of the differential comparison of inter and intera specific craniometric comparison of family Equidae. The most extensive craniometrical investigations consisted of 56 measurements of different parameters of *Equus* skull. The parameters of variable features were recorded as akrocranion-prosthion (483.8  $\pm$  48.2 mm), nasion-prosthion (264.2  $\pm$  28.7 mm), facial width (161.6  $\pm$  13.9 mm), Ectorbitale-Ectorbitale (179.6  $\pm$  19.2 mm), Basion-prosthion (421.2  $\pm$  53.0 mm) and Nasiontermaxillare-prosthion (205.0  $\pm$  63.6 mm). All the features have been reported as mean  $\pm$  standard deviation. The significant differences (p<0.005) were found in the skull parameters while compared inter-specifically, when one way ANOVA was applied while no significant differences were found intera-specifically when ANOVA was applied on features of adult individuals. Thus, on the basis of craniometrical assessment we assigned two species viz. *E. caballus* and *E. asinus* to the studied samples. The current study reports the comprehensive craniometrical analysis of *Equus* from Pakistan which adds the data regarding the shape and size of *Equus* skull in global craniometrical datasets of family Equidae.

# RESISTANCE OR SUSCEPTIBILITY OF EIGHT AUBERGINE CULTIVARS TO MELOIDOGYNE JAVANICA

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Variations were observed in resistance or susceptibility among eight aubergine cultivars to *Meloidogyne javanica*. Brinjal Jamak was the only cultivar found to be moderately resistant. Two cultivars namely Brinjal Shilpa and Singh Nath 666 appeared moderately susceptible. Five cultivars *viz*. Round Black, Short Purple, Brinjal PPL, Global Brinjal PPL and Namyal Ratchburi behaved as susceptible. All the cultivars behaved differently regarding formation of galls, egg masses, number of eggs per egg mass and reproductive factor. Maximum galls, egg masses, eggs per egg mass and reproductive factors were observed on Round Black followed by Global Brinjal PPL and the minimum were recorded on cultivar Brinjal Jamak. Similarly, significant effects of *M. javanica* were observed on growth parameters of these cultivars. The reductions in moderately resistant cultivar were significantly lower as compared to the moderately susceptible and susceptible cultivars. The maximum reductions in shoot and root lengths and shoot weight were recorded in case of Round Black followed by Global Brinjal PPL. On the other hand, the minimum reductions in these parameters were found in Brinjal Jamak. Similarly, the infection of *M. javanica* caused an increase in root weights of all the cultivars. The increase in root weight was the minimum in cultivar Brinjal Jamak while it was the maximum in case of Round Black followed by Global Brinjal PPL. Regression analysis

showed positive and significant relationships between number of galls and reductions in shoot and root lengths and weights. As the plants of moderately resistant cultivar Brinjal Jamak suffered less damage and suppressed nematode infection considerably and therefore, recommended for cultivation in root-knot nematode infested fields to abate yield losses and repress the nematode from further multiplication.

# HISTOPATHOLOGICAL CHANGES IN THE SPLEEN INDUCED BY NEMATODE PARASITES IN CATFISH, *ARIUS SERRATUS* (DAY, 1877) FROM KARACHI COAST

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In the present study the histopathological changes in the spleen of catfish, *Arius serratus* (Day, 1877) from Karachi coast due to the nematode parasites is described. For sampling purpose, fish were purchased from Karachi fish harbor and brought to the parasitology laboratory for further detailed investigation during April, 2019. Histological sections were prepared by using standard procedures of parasitology. Selected liver tissues were fixed and stained by using eosin and haematoxylin. Eight *Raphidascaris acus* (Bloch, 1779) larvae were found attached to visceral mesenteries, liver and spleen of *Arius serratus* (Day, 1877). The histopathological changes include morphological changes, necrotic abnormalities, inflammatory response of the host tissues, enlarged cells, disintegration and degeneration of spleen cells and tissues were observed.

# RISK FACTORS ASSOCIATED WITH DENGUE PREVENTION AND CONTROL IN TEHSIL KASUR PUNJAB PAKISTAN

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Dengue fever (DF) appears to be emerging in all districts of Punjab, Pakistan in the recent year from January to December 2019. Our under-study area is tehsil Kasur. A preventive case control study was performed to investigate risk factors for the development of DF in tehsil and district Kasur. A total of 43 union councils having patients with DF were studied in that area. Out of 43 union conceals, 11 belongs to urban area of city Kasur and two are urban Mustafa Abad, one is from urban Khudian Khas and one is urban from Raja Jung and remaining 27 belongs to ruler area of tehsil Kasur. The risk factors for this under studied area abut dengue patients and larvae positive are open sewerage system, unhygienic conditions found during outdoor surveillance in urban area street canals fund stagnant water especially in tannery area and other under construction buildings and living in a house discharging sewage directly into to ponds were all significantly associated with larvae breeding sites. These all factors provide best habitat for the growth of dengue larva at that area. These results contribute to the understanding of the dynamics of dengue transmission in tehsil Kasur and its vicinity, which is needed to implement dengue prevention and control programs effectively and efficiently.

# PREVALENCE OF THEILERIOSIS AMONG CATTLE IN DISTRICT LAKKI MARWAT, KHYBER PAKHTUNKHWA, PAKISTAN

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Theileriosis is a tick-borne haemoparasitic disease and is responsible for huge economic losses in livestock sector of Pakistan. The present work was carried out to determine the prevalence of Theileriosis among cattle in

district Lakki Marwat, Khyber Pakhtunkhwa, Pakistan. A total of 138 blood samples of cattle were collected during the period from January 2018 to June 2018 and were examined microscopically for theileriosis infection. In 138 blood samples, 17.39% samples were found positive with theileriosis infection. The overall prevalence in male cattle (18.36%) showed that theileriosis is more prevalent in male cattle as compared to female cattle (16.85%). Prevalence among cattle was higher in age groups of less than one year (20%) as compared to more than one year of cattle (16.85%). Month-wise prevalence of theileriosis was recorded higher in June (33.33%) while no infection of theileriosis was found in January and February. It was concluded that theileriosis is prevalent in the district Lakki Marwat, Khyber Pakhtunkhwa, Pakistan. It is highly recommended that there is a need for further epidemiological investigations using advanced serological and molecular techniques.

# ECO-FRIENDLY APPLICATION OF MEDICINAL PLANTS TO CHECK THE BIOTOXICANTS AND REPELLENT EFFECTS ON PRIMARY STORED GRAIN INSECT PEST *TRIBOLIUM CASTANEUM* (COLEOPTERA: TENEBRIONIDAE)

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Red flour beetle (*Tribolium castaneum*) is very destructive pest of stored grain and cereal in hot and dry climates of the world. The present study was conducted to check the insecticidal efficiency and repellency of different plant extracts against the larvae of red flour beetle which are environmentally safe and have no mammalian toxicity. The experiment was conducted at Entomology Lab, College of Agriculture Bahauddin Zakariya University, Bahadur Sub-campus, Layyah under lab conditions, with CRD design. The experiment was repeated three times. The plant extract of fennel flower (*Nigella sativa*), turmeric (*Curcuma longa*) and wild rue (*Peganum harmala*) were used with three concentrations 3%, 6% and 9%. The data was collected after 24-hour post treatment. The minimum percent mortality was recorded after 24-hour exposure period i.e. 33.33, 6.667 and 8.33% on the concentration of 3, 6 and 12% of turmeric as more mortality was observed in fennel seed as compared to turmeric. The maximum mortality (35.00%, p≤0.01) of red flour beetle was recorded at highest concentration at 12% followed by 10.71% at minimum concentration 3%after 120 hour of exposure period. The overall results showed that corrected mortality was increased with increase of time and concentration.

# EFFICIENCY OF RED SEAWEEDS AGAINST DENGUE VECTOR MOSQUITO, AEDES AEGYPTI (L)

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Marine plants occupy two-third of the earth's surface and are an untapped reservoir of the novel compounds. The role of the seaweed natural product in drug discovery is prodigious, especially for the development of anticancer, antiviral and antibacterial. Therefore, in the present study, a particular attempt has been made to assess the toxicological effects of seaweeds against the dengue vector mosquito, *Aedes aegypti* (L). Dengue is the most prevalent vector-borne viral disease and has affected nearly half of the world population. Its adult is also the primary carrier of Chikungunya, and Zika virus. In this experimental work, four species of red seaweeds *Laurencia karachiana sp. nov, Gracilaria foliifera, Jania rubens, Asparagopsis taxiformis* were collected from the Karachi coast and extracted with hexane, dichloromethane and methanol. The efficiency of red seaweeds extracts against *Ae. aegypti* (larval, pupal and adult) stages were determined by using dose-response bioassay method. Separate investigation on the toxicity and IGR effects were carried out. The comparative study showed that the hexane

extracted from *Jania ruben* exhibited potent toxic effect ( $LC_{50} = 32 \square g/mL$ ) after 24 h of treatment. The high knockdown effects were also noted after 1-6 h of treatment. The chitin synthetic inhibiting, morphological abnormalities, and growth inhibiting (IGR) effects were noted in the median and higher concentrations of the dichloromethane and methanol extracts resulted in the formation of immature such as larvi-pupae form. Study showed red seaweeds, occupying Arabian Sea possess anti insect property that can be used as green pesticides.

### ANTHROPOGENIC IMPACTS ON AVIAN AND MAMMALIAN DIVERSITY IN MARGALLA HILL NATIONAL PARK, ISLAMABAD-PAKISTAN

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This study was designed to know the diversity with the avian and mammalian species. The data collected through direct and indirect methods. During survey, total 153 avian and 26 mammalian species were recorded from Margalla Hills National Park. Simpson's index (S) provides quantitative report of avian diversity which was highest at forest (0.9888) followed by urban (0.8995), rural (0.974) and agriculture (0.9529). Similarly, Simpson's index provides quantitative report of mammalian diversity which was highest at forest (0.8615) followed by Urban (0.6593), Rural (0.6852), Agriculture (0.4518). This study showed that urbanization and agriculture intensification were negatively impacts on the avian and mammalian diversity.

# HUMAN ATTITUDES TOWARDS HERPETOFAUNA AMONG THE PEOPLE IN AZAD JAMMU AND KASHMIR-PAKISTAN

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Questionnaire survey was distributed to 1000 people in the state of Azad Jammu and Kashmir, Pakistan, belonging from different district. The main aim was to obtain data regarding the hypothesis that human-associated persecution is directly influence by the existence of wrong ideas and negative values. A Principal component analysis method was used to confirm the hypothesis about the possible relationship between negative values and observations, and anti-conservation and persecution attitude towards reptiles and amphibians. Scio-demographic variables were also added. The results clearly suggest that folklore and negative values noticeably predict anti-conservation attitude toward herpetofauna.

# ASSESSMENT OF HERPETOFAUNA DIVERSITY IN MUZAFFARABAD, AZAD JAMMU AND KASHMIR, PAKISTAN

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The main objectives of the study are as; to estimate the diversity and density herpetofauna in the study area. The data were analyzed through direct and indirect counts. Binocular (32x50) was used to observe the animals. During the research noted that having, 2 order, 7 families, 14 genera and 18 species recorded from Muzaffarabad. It is noted that *Bufo himalayanus* is most common (R. A= 0.13) species of the area. While it is noted that *Eublepharis macularius* is lowest species (R.A, 0.02) species of area.

# ASSESSMENT OF HUMAN ACTIVITIES IMPACT ON AVIAN AND MAMMALIAN DIVERSITY IN AYUBIA NATIONAL PARK, KHYBER PAKHTUNKHWA-PAKISTAN

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The avian and mammalian species has threats due to human activities and interactions. Therefore this study was designed to know the anthropogenic impact and interaction with the avian and mammalian species. The data collected through direct and indirect methods. Statistical analysis is done with Past Software. During present survey, a total of 117 avian and 21 mammalian species were identified from Ayubia National Park. During the study noted that highest Shannon-wiener diversity index was recorded from forest (4.393) as compared with other habitats urban (2.061), rural (3.771) and agriculture (3.573). And Shannon wiener index provides quantitative report of diversity which was highest at forest (2.441) followed by urban (1.871), Rural (1.665) and agriculture (1.362). The statistical analysis showed that anthropogenically disturbed habitats negatively impact on the diversity of avian and mammalian diversity.

# POPULATION AND CONSERVATION STATUS OF BARKING DEER (Mantiacus mantjac) IN THE MARGALLAH HILLS NATIONAL PARK, ISLAMABAD

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To assess the current population and conservation status of barking deer (*Muntiacus mantjak*) a study was conducted from July to December 2019 in the Margalla Hills National Park (MHNP) Islamabad which falls in Himalayan foothills between 450m-1500m elevation. The data were collected by direct (observations and signs) and indirect (questionnaires) methods on seven selected sampling sites of the study area. Field observations were taken by using line transect method. The study area was divided into seven sampling sites: village Rumli, village Ratta hottar, Trail-3, Trail-4, Trail-5, Trail-6 and Saidpur. The sampling was done at dawn and dusk while day time was utilized for taking indirect data. An average population density of 2.89 per km² deer was estimated with the highest population at Trail 3 (3.42/km²), followed by Trail 5 (3.02/km²), Trail 6 (2.72/km²), Trail 4(2.57/km²), Rumli (2.38/km²), Ratta Hottar (2.17/km²) and Saidpur (1.86/km²). The maximum population density was recorded at on Trail 3 3.42 per km² while the lowest density was recorded at the Saidpur 1.86 per km². The study revealed a sharp boost up in the population of barking deer in the (MHNP) due to conservation efforts made by Islamabad Wildlife Management Board, Islamabad. Although, forest damage, hunting, pouching and over grazing by livestock were the major threats to the conservations of barking deer in the study area.

# ASSESSMENT OF TOXIC EFFECT OF ARSENIC ON GROWTH AND HAEMATOLOGICAL PARAMETERS OF *LABEO ROHITA* DURING SUB-LETHAL EXPOSURE

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The main objective of present study was to determine the toxicological effect of arsenic at sub-lethal concentrations on growth and haematological parameters of *Labeo rohita*. After acute toxicity trials, *Labeo rohita* 

were exposed to sub-lethal concentration of arsenic and at intervals of 14 and 28 days, the growth and haematological parameters were elevated. Growth performance was significantly higher in unstressed fish as compared with arsenic exposed *Labeo rohita*, with final weight gain (5.4±0.08g and 4.4±0.17g), (7.4±0.21g and 5.4±0.16) at 14 and 28 days, respectively. Growth reduction is strongly correlated with arsenic toxicity. After chronic exposure of arsenic, blood of fish was taken and processed for estimation of haematological parameters. The hematological analysis of arsenic exposed *Labeo rohita* at 14 and 28 days exhibited significant decrease in red blood cells count (RBCsC), hemoglobin (Hb) and hematocrit (Hct) content while noticeable elevation in white blood cells count (WBCsC), mean cell volume (MCV), mean cell hemoglobin (MCH) and mean cell hemoglobin concentration (MCHC) was recorded in comparison to control. According to this data, it is clear that arsenic is highly toxic for fish. The altered parameters can be used as biomarkers for arsenic detection in aquatic environment.

#### PLANKTONIC DIVERSITY IN THE WATER OF RIVER RAVI

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Phytoplanktons are usually microscopic organisms, versatile or generally greenish in coloration with various species classified on the basis of their distinctive shape and size. Few species play pivotal role in fish survival and human consumption in raw form in various parts of world. Main feature is that they are the main bio indicators of freshwater as well as oceanic environment. The present study entitled "Determination of planktonic diversity in river Ravi" was carried out at Ravi embankments and center near Shahdara, Ravi Bridge, Lahore. Severely polluted water with industrial effluents and sewage containing minute phytoplankton species such as *Chlamadomons reinhardtii*, *Microcystic, volvox, scenedesmus sp., Pediastrum duplex, Cladophora sp., Closterium* along with 7 species of zooplanktons.10% solution of formalin was used for the preservation of both types of plankton (zooplankton and phytoplankton). Light Microscope was used for the identification of the preserved plankton. Images were taken with the help of camera. Observed planktons are important as a natural fish feed consumed by freshwater fishes such as grazers and suckers summarily dependent upon planktons and their respective general orders and suborders.

#### STUDY OF PLANKTONIC DIVERSITY FROM THE WATER OF RIVER JHELUM

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Planktons are water flooding organisms which swim only in the direction of water current. There are two main types of plankton, phytoplankton and zooplankton. Fish directly depend upon plankton for feed. So, plankton play key role in the food web in the water ecosystem. The present study was conducted from the water of River Jhelum for the diversity of planktons. River Jhelum play essential role in economy of Pakistan with regarding to agriculture. River Jhelum started from northwest side of Peer Panjal and flows to the parallel of Indus River. In order to measure and evaluate planktonic community we take the water sample from River Jhelum. From this water sample we take out plankton by using net and identified them with the help of light microscope. Some of Phytoplankton included Closterium and Diatom. Zooplankton included Cyclops, Chalamydomonas, Ceriodaphnia, Amoeba, and Paramecium. Beside this it is important point that plankton diversity increases and decrease with fluctuation in environmental condition. But due to anthropogenic activities pollution level is increase in River Jhelum that negatively affect to planktons.

# ISOLATION, CHARACTERIZATION AND ANTIOXIDANT POTENTIAL OF BIOACTIVE COMPOUNDS FROM MORINGA OLIEFERA

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Moringa oliefera is the most versatile and fast growing plant, belongs to the family "Moringaceae". Every part of this plant is important source of terpenoids and phenolic compounds, methionine, cysteine, Fe, vitamin A, B, C, and essential sulfur containing amino acids. The aim of this work is to review the use of M. oliefera due to its nourishing and antioxidant qualities. In this research the extract from leaves of M. oliefera were used to determine total phenolic contents by using Follin-ciocalteu reagent. Characterization of the phenolic compounds present in the extract was done through HPLC. Phytochemical analysis was done to find out the presence of Flavonoids, Alkaloids, Saponins, Tannins and Quinones. Antioxidant activity was examined by using the DPPH radical scavenging test. Antioxidant properties were also observed while using different enzymatic protocols (superoxide dismutase, peroxide dismutase, catalase, protease and  $\alpha$ -amylase). The activity of plant extract against various bacterial and fungal strains was evaluated along with Minimum Inhibitory Concentration (MIC). In the last cytotoxicity of the Moringa extract was observed by assessing the hemolytic activity against red blood cells of human being. Outcome of this research offers support to the previous works mentioning the usage of M. oliefera for edible and therapeutic applications.

# THERAPEUTIC POTENTIAL OF SEED EXTRACTS FROM SEVEN DIFFERENT MEDICINAL PLANTS

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In the present study we examined seven plant seed extracts including Momordica charantia (bitter melon), Vitis vinifera (grape), Citrullus lanatus (water melon), Moringa oleifera (drumstick tree), Punica granatum (pomegranate) Psidium guajava (guava). and Cucurbita maxima (pumpkin). Twenty five components of oil were determined by the evaluation of oil through GC/MS, that consisted upto 90.9% of the seed oil. The major components were trans- nerolidol apiole, germacrene and cis-dihydrocarveol. Most abundant component in seed oil was alpha- eleostearic acid, that has several therapeutic applications of decreasing the fat contents in blood, lessens cardiovascular diseases, reduces the proliferation of cancer cells resulting in anticancer activities and antiinflammatory effects. Polyphenolic constituents were present in large amount in V. vinifera and its total phenolic content depend upon the seed type. The substances having Vitamins (C ,A ,E ,B1 ,B2 ,B3 as folate), cell antioxidants,, minerals( potassium, calcium, magnesium, zinc, phosphorous and iron) and different phytochemicals ( such as saponins, terpenoids, glycosides, phenolic acids, resins alkaloids reducing sugars, proteins and free acids) were available in M. oleifera and M. charantia, also its extract could be used as remedy for diabetes. The chemical components isolated from P. granatum were hydroxybenzoic acids that included ellagic acid, 3- di-o-methyl ellagic acid, 3,3,4-tri-O-mrthylellagic acid, conjugated fatty acids and punicic acid. Due to phenolic composition, the seeds manifested anticarcinogenic, gastrointestinal and antimicrobial activities. There was significant reduction in levels of cholesterol, glucose, triglycerides and body weight when the Wistar rats, when treated with P. guajava seeds. Different flavonoid and phenolic compounds were isolated. In the kernels 27% unsaturated fatty acids, 66% saturated fatty acids, 11% stearic acid and 16% palmitic acids were present, while 13% oleic and 53% linoleic acid were also found.

# FEEDING BEHAVIOUR OF SOME MANTIDAE SPECIES (MANTODEA) FROM MATIARI, SINDH, PAKISTAN

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The sum of activities of animals that is directed towards the procurement of nutrients is collectively called as feeding behaviour. The same food or the prey is procured differently by different predators with the help of special capabilities i.e Formicidae and Mantodea. Mantodea is a group of mostly large and conspicuous predatory insects with versatile, unique and special capabilities. They prey upon a wide array of animals, ranging from springtails to small vertebrates hence are the best biological pest control agent. Observing the feeding behaviour of praying mantids under natural conditions is difficult due to their speed, camouflage, low lying in vegetation etc. That's why this study was under taken to see feeding behaviours of three diverse occurring species in Laboratory conditions. Consequently their oothecae (egg case) were collected sorted out for raring. The collected oothecae fixed in aerated cadges with all required factors and after hatching the feeding behaviours of *Tenodera attenuata*, *Sphrodomantis transcausica* and *Mantis religiosa* was recorded.

#### BIODIVERSITY OF SPIDER FAUNA FROM VARIOUS DISTRICTS OF SINDH, PAKISTAN

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Sindh is the third largest province of Pakistan (Karachi is its capital), extended 579 km from north to south and 442 km from east to west, with an area of 140,915 km is subtropical agricultural zone (has four weathers) having appropriate to complimentary average temperature conditions which give positive favoritism to biota. Due to this a variety of crops are grown, and people of this area frequently use pesticides, to control pests, but extra they bestow dangerous consequence on many non-target animal fauna including spiders. Spiders are macro invertebrate predators in agro and non agro ecosystems and habituated in every part of terrestrial environment. They can drastically minimize pests in agricultural fields and are best bio-control agents. In support of this, biodiversity of spider fauna undertaken to see the species richness and evenness while measured with three Simpson's Indexes. The spiders were collected from agro and non agro fields and organize an inclusive and updated record of biodiversity of Arachnida occurring in 20 districts of Sindh. Throughout survey 596 spiders were collected and arranged into 9 families, 28 genera and 59 species. We found supplementary spider fauna in uncultivated fields than cultivated fields.

# DIVERSITY AND ECOLOGY OF ANTS (FORMICIDAE) FROM NAUSHAHRO FEROZE, SINDH, PAKISTAN

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The study was carried out from District Naushahro Feroze (located 26° 50′24″ N 68° 07′ 12″ E) with altitude 38 meters. Diversity of ant's fauna occurring in an ecosystem has diverse and versatile importance. Because of their huge number and functions like soil fertility, predacious, scavenger, pollinators and pest exterminators. They are social insects living in colonies having different cast i.e workers, drone, queen and soldiers. six colonies were studied and total 50 specimens of each colony (with all castes) were collected and preserved into 75% ethanol with few drops of glycerine. Collections were made by hand picking and using bait (sweets and chicken visceral) and arranged into

fifteen species and five genera. Identification was prepared using keys given by Bolton, 1994; Sheela, 2008; Naumann, 1993 and McArthur, 2001. Pictures were captured by Stereoscopic microscope with LED and digital Camera.

# BIODIVERSITY AND MORPHOLOGY OF ANTS (HYMENOPTERA: FORMICIDAE) INHABITING ON MANGO TREES FROM HALA, SINDH, PAKISTAN

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Biodiversity means variety of life while morphology describe as external physical appearance, here both term are used for ants. Association of ant fauna is vital for agriculture. Like earth worm is famous for the soil fertility, bees with pollinations, mantids and spiders with pest control, house fly cleanses of debris etc in the same way the ants perform all roles (Predators, Scavengers, Pollinators, Soil turners mean soil fertility and sometimes pests also) in the ecosystem. They are social insects having a variety of colors. This study is based on the biodiversity of ants associated with mango trees grown in Tando Allahyar. Collection was made from mango tress using bait like chicken visceral, sweets and insects while putting this bait on white paper sheets. Total 2096 specimens were collected and sorted out into six species and five genera. Identification was done keys given by Bolton, 1994 and Sheela, 2008. This is the first reported research work from Sindh, Pakistan.

# CHARACTERIZATION OF SCORPION SPECIES COLLECTED FROM BHIMBER, AZAD JAMMU AND KASHMIR

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Scorpions are among the oldest terrestrial arthropods, which are distributed worldwide, except Antarctica and some pacific islands. The study sites were selected based on different topographic condition such as foothill and mountains. Daytime collection was carried out using hand digging tools for turning stones and excavate the burrows, as well as night samplings were done by using ultraviolet light, the collected specimens were maintained in 70% Ethanol. Morphological characterization was done on the basis of color, body shape, prosomal, mesosomal and metasomal characters. Molecular characterization was done on the basis of CO1 gene. A total sixty four specimens were collected including juveniles, young and adults. Out of which six were *Hetrometrus latimanus* and 58 were *Hottentota tamulus*. *Hetrometrus latimanus* was collected from stony land while *Hottentota tamulus* were collected under the small rocks with clay. DNA Barcode analysis showed they are closely related to already described species of respective genera.

### TETRACONODON AND SIVACHOERUS (SUIDAE) FROM POTWAR PLATEAU, NORTHERN PAKISTAN

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Rare tetraconodonts, *Tetraconodon magnus* and *Sivachoerus prior* are ascribed in this article. *Tetraconodon* is believed to be occurred in the Upper Dhok Pathan and Tatrot formations of the Siwalik Group. However, the

specimens described here from the mid Dhok Pathan Formation are important in relieving the long-held notion of the previous researchers. Similarly, *Sivachoerus prior* appeared earlier than it was thought previously.

# NEW REMAINS OF *DORCATHERIUM* (TRAGULIDAE) FROM MIDDLE MIOCENE OF CHAKWAL, PAKISTAN

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In current study, the new dental material collected from three sites of Chinji Formation, are described and discussed. The recovered material includes the cheek teeth including deciduous as well as permanent. The morphology and size of studied specimens here match with two *Dorcatherium* species *D. minus* and *D. majus*. The aim of study is to enhance knowledge on middle Miocene *Dorcatherium* specially to document the deciduous premolar of *Dorcatherium* which are least known.

# MORPHOMETRIC VARIATIONS OF THE SKULL IN THE GREY WOLF (CANIS LUPUS) IN PAKISTAN

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The current study reports the morphological variation in the skull of grey wolf (*Canis lupus*). The grey wolf (*Canis lupus*) belongs to the family Canidae and is a widely distributed in all of the mountainous ranges of Pakistan, from Baluchistan up to Chitral and Gilgit Baltistan in the north. Total 10 skulls of grey wolf were analysed for 24 cranial variables which have been measured from the dorsal and ventral side. Six indices of each skull were also generated by using the primary variables to analyse any shape variation among the skulls. All the variables were analysed with the help of SPSS software by using student's t-test to determine statistical significance at significance level (p<0.05). The significant differences (p<0.05) were found (p<0.05) which indicates intera-specific variations among the studied individuals. The observed morphometric variation exists in relation to shape and size of the skulls. The study reports the first morphometrical analysis of the skull of grey wolf from Pakistan and adds significant data in the available global data set of family Canidae. Furthermore, molecular bar coding characterization may confirm the specific diversity of grey wolf in Pakistan.

# FIRST RECORD OF *HYSTEROMORPHA* TRILOBA, RUD, 1819 IN BLACK-CROWNED NIGHT HERON *NYCTICORAX NYCTICORAX* FROM HAMAL LAKE, SINDH, PAKISTAN

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A detailed assessment was done on BLACK-Crowned Night Heron *Nycticorax nycticorax* to explore parasitic forms of helminths from 10.2016 to 09.2017. Twenty four Black-Crowned Night Herons were thoroughly

investigated after dissection. All dissected *Nycticorax nycticorax* were found with heavy load of different forms of helminth parasites such as trematodes and ssnematodes. Having done dissection and teasing of all visceral organs, they were carefully observed on stereo dissecting microscope. The drawings of the recovered specimens were made with the help of microscope attached with drawing tube and photographs were taken with Omax Camera attached trinuclar microscope. 09 out of 24 Black-Crowned Night Herons were found positive with *Hysteromorpha trilobal*, Rud, 1819. Sixty eight specimens of the parasites were recovered from 09 hosts. The recovered specimens of the parasites were identified with help of different keys, internet and supervisors. The species found in the present research project is the first record from Sindh, Pakistan.

# CURRENT STATUS AND DISTRIBUTION OF *EUPHLYCTIS CYANOPHLYCTIS* (COMMON SKITTERING FROG) AND *BUFO STOMATICUS* (MARBLED TOAD) IN KARACHI DISTRICT, SINDH.

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The present work was carried to determine the current status and distribution of two species of order anura i.e., Euphlyctis cyanophlyctis (Common Skittering Frog) and Bufo stomaticus (Marbled Toad) in Karachi district, Sindh. During the study, both species were recorded throughout the year 2019 in selected areas of Karachi district. It was found that Euphlyctis cyanophlyctis was rated as least concern anuran whereas Bufo stomaticus was found to be abundant and its population can be considered as stable in Karachi district. On the basis of present study, it is concluded that the population of anurans is altered due the impact of anthropogenic activities which include urbanization, improper disposal of industrial and domestic wastes and overutilization of pesticides and chemical fertilizers in the agricultural fields. Other factors include climate change, habitat destruction and lack of sufficient rainfall in Karachi district.

# COMPARATIVE EVALUATION OF DIFFERENT HONEY TYPES FROM A. MELLIFERA HONEYBEES OF PAKISTAN

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A study was conducted at Honeybee Research Institute, National Agricultural Research Centre, Islamabad in 2017 to evaluate the physicochemical characteristics such as moisture, free acidity, pH, electrical conductivity and total sugars of 19 honey samples collected from different floral sources and floral origins. The purpose was to evaluate the physicochemical characteristics such as moisture, free acidity, pH, electrical conductivity, total sugars and microscopic analysis of 19 honey samples collected from different floral sources and floral origins. The range of physicochemical parameters for Margalla brand and other branded honey samples was 17.96±0.25 to 23.23±0.25 % for moisture, 8.67±1.53 to 25.50±0.50 meq/kg for acidity, 0.15±0.01 to 0.96±0.02 mS/cm for electrical conductivity, 3.77±0.03 to 6.86±0.02 for pH and 75.33± 0.29 to 80.60±0.17% for total sugars. Non-branded honeys had moisture in the range of 17.76±0.25 to 23.96±0.25%, 8.00±1.00 to 29.91±0.38 meq/kg acidity, 0.13±0.01 to 0.71±0.01 mS/cm electrical conductivity, 3.75±0.02 to 6.92±0.02 pH and 74.46±0.25 to 81.33±0.29% total sugars. All the honey samples were found to meet Codex Alimentarius and European Union Directives standards except for the moisture content, pH and electrical conductivity in some samples. Sider honeys showed significantly higher values of pH and Electrical conductivity and lower values of acidity as compared to the other honey types. The results reported in this study can help the beekeepers, industrialists, researchers, and consumers for better understanding the honey quality and properties.

#### EFFECT OF LACTOBACILLUS, ANTIBIOTICS, AND NANOPARTICLES TO STAINS OF PSEUDOMONAS AERUGINOSA FROM ACUTE AND CHRONIC PULMONARY INFECTIONS

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Antibiotic resistance in *Pseudomonas aeruginosa* is a major barrier to successful treatment of infection. Additional and novel measures to control this pathogen are needed, along with contemporary information about antibiotic resistances that are present in isolates from different environments. In the present study 72 samples from blood, 43 from sputum, and 19 were obtained from tracheal aspirates patients suffering from chronic and acute lung infections admitted to a local hospital in Lahore. Susceptibility of 134 isolates of *P. aeruginosa* was tested against selected antibiotics (meropenem, imipenem, piperacillin, amoxicillin, amikacin, gentamicin, tobramycin, clarithromycin, cefepime, cefixime, levofloxacin, and ciprofloxacin), Lactobacillus strains and metal nanoparticles (copper, ferric and zinc). *P. aeruginosa* isolates showed in vitro resistance against 11 of 14 antibodies tested. The isolates were highly susceptible to meropenem, piperacillin, and amoxicillin. It was also observed that the growth of these resistant *P. aeruginosa* strains was significantly inhibited in the presence of Lactobacilli *spp.* and nanoparticles of silver, zinc and ferric oxide at a concentration of 12, 200 and 1µg/ml, respectively. This study may help in the development of chemotherapeutic methods against multidrug resistant bacterial pathogens in chronic and acute lung infections. It provides a practical approach towards the use of nanoparticles to enhance antimicrobial activity against these pathogens.

# ASSESSMENT OF NEMATODE INFECTION IN FRESH WATER FISHES OF INDUS RIVER, PAKISTAN

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A total of 792 specimens belonging to 29 species of fishes were examined and 24.24% were infected with nematodes. The highest prevalence of nematode observed in *Aorichthys seenghala* (55.55%), followed by *Rita rita* (43.90%), *Channa mariulius* (43.33%), *Wallagu atta* (41.46%), *Xenentoden cancila* (41.37%), *Oreochromis niloticus* (38.88%), *Cyprino carpio* (28.57%), *Eutropiichythys vaucha* (27.77%), *Oreochoromius mossambius* (25%), *Mastacembelus siamensis* (25%), *Channa straita* (24.24%) *Mastacembelus armatus* (21.87%), *Mystus armatus* (21.87%) *Heterpneustus fossils* (22.585) *Ompok pabo* (20.80%) *Mystus vittatus* (18.18%) *Catla catla* (16.66) *Labeo gonius* (10.34%) *Mystus caviasus* (9.75%) and no infection noticed in *Notoperus notopterus*, *Cirhinus mirgla*, *Cirhinus cirrhosus*, *Cirhinus reba*.

#### PROLIFERATION OF THE SPIDERS IN LABORATORY

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It is known worldwide that there are more than 27,000 species of spiders. There are about 700 species of spiders in Uzbekistan and 18 of which have neurotoxic effects on human being. A number of scientists, led by N. E. Ergashev, conducted research works on the reproduction of biologically active spiders in laboratory conditions at the Institute of Zoology

of the Academy of Sciences of the Republic of Uzbekistan. The purpose of this work is to study some of the characteristics of the proliferation of the spiders in laboratory. The study examined the habitat and behavior of the spiders of Allahogna singoriensis (Lachmann, 1770) where abiotic indicators (temperature, humidity, food, etc.) were created and maintained in captivity collected in the Syrdarya region. Flour bug (Tenebrio molitor molitor L.) was breeder and used as a main food for spiders. According to the results of the study, A. singoriensis caught in the Syrdarya region, started the cocoon in December. In the nature, spiders begin laying eggs in April-May. The off springs emerge from the cocoon immediately reach to the top of their mother after 7-8 days their start live independently. It was studied that they put 3-4 cocoons in captivity and there are 150-200 spiders in each cocoons. Last autumn, In Sharof Rashidov district of Syrdarya region, it was found that out of 36 spiders (A.singoriensis), 13 of them (8) and 4 (5) produced the cocoon in captivity. After the first cocoon, it was recorded that the second one produced cocoon in 39, the third one in 44, and the fourth one in 47 days. There were no spiders in the fourth cocoon. As it was observed that after 34 days from the start of cocoon, off springs at age of 1 emerge on the mother's abdomen immediately. It was found that the mother carries her offspring on the shoulders for an average of 10-12 days. Thus, it was obvious by observation that spiders have no problem on breeding. It should be noted that breeding process of spiders in captivity do not diminish their activity during early spring, late autumn and winter, and live closer to their natural habitat. Spiders build their nest in the sand which is put into the special glass dishes. As a main food for young spiders 1-2 year old bran bugs are given, and adult spiders are given beetle. Research has shown that spiders can produce several generations throughout the year if the temperature is at 26.5° C and humidity is at 54 % in the winter and autumn, and provide adequate nutrition.

# BPA INDUCED CANCER AND NEUROMUSCULAR DEGENERATION MODELLING IN DROSOPHILA MELANOGASTER

#### Anusha Amanullah, Sharon E D'Souza, Ayesha Ashraf Baig, Ifrah Mehmood, Sanya Shabbir and Mushtaq Hussain\*

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Bisphenol-A is a compound extensively used in the manufacturing of plastics. In recent times, several studies have pointed its toxicological effects on human health. *D.melanogaster* is a model organism for studying genetics, developmental biology and human diseases including cancers and neuromuscular degeneration. The primary aim of the present investigation is to explore the effect of Bisphenol-A, a compound used mainly in the plastic industry, on the brain development in *D. melanogaster* fruit fly. Briefly, *D. melanogaster* flies inbred line were exposed to Bisphenol-A by dividing the flies into five sets: control group, 1mM, 2mM, 4mM and 10mM Bisphenol-A. At the end of the experiment, pupariation and fecundity of the F1 progeny flies were assessed. Flies from each groups were compared for morphological traits. Similar anatomical investigation was focused on brain and thoracic muscles. Exposure of the Bisphenol-A resulted in the noticeable change in various traits compared to the control set (standard cornmeal medium). Variations in the number of offspring suggest that Bisphenol-A affect the reproductive functions of the flies. Similarly, noticeable changes in anatomy and/or gene expressions pointing to the gene expressional aberrations leading to cancers and neuromuscular degeneration.

#### WHOLE GENOME DUPLICATION AND EVOLUTION OF CYLD GENE IN VERTEBRATES

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CYLD, cylindromatosis, gene encodes an ubiquitin specific proteases that stabilize protein linked with an array of biological role, NFkB mediated inflammation, p53 and RIG mediated apoptosis. The present investigation aims to explore the evolution of CYLD gene in vertebrate lineage underlying genomic mechanism. Briefly, genomes of

species of vertebrate lineages were explored for the presence of CYLD gene homologues. The retrieved sequences were aligned for C19 domain and phylogenetic tree was reconstructed with 1000 bootstrap replicates employing JTT+G evolutionary model. Phylogenetic tree was then realigned with genomic physical map of genes, gene structure and protein domain architecture. Finally, full length molecular models of ancestral form of CYLD, CYLDA and CYLDB were constructed and compared for structural attributes. Genome mining demonstrates variable number of homologues in different species of vertebrates with mammals have only one CYLD homologue whereas fishes have maximum of six homologues of respective gene. Phylogenetic analysis divided all sequences into two superclades, CYLD I and CYLD II. CYLD I comprises three major clades, CYLDA, CYLDE and CYLDC. Whereas, CYLD II coalesced two clades, CYLDB and CYLDD. Distribution of species amongst clades and superclades points that first two rounds of whole genome duplication at the base of vertebrate led to the origin of four CYLD paralogues, CYLDA, CYLDB, CYLDD and CYLDE. In comparison CYLDC, CYLDF and CYLDG are the product of third and fourth round of whole genome duplication in fish lineages. Primary and tertiary structural comparison of paralogous proteins and site specific selection pressure points to the neofunctionalization and/or sub functionalization among different CYLD paralogues. Findings of the present study are the first to resolve the evolutionary roadmap of CYLD at genomic resolution. Thereby covering the lacuna in the understanding of CYLD biology. The study is supported by the Higher Education Commission

### COMPARATIVE SUSCEPTIBILITY AND LIFE HISTORY OF CALLOSOBRUCHUS MACULATUS F. ON VIGNA UNGUICULATA STORED SEEDS

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Callosobruchus maculatus F. is a serious pest of stored grains, Such as green gram, cowpea, black gram and chickpea. 100g of different Cowpea varieties were taken to infestation and damage by the storage beetle, C. maculatus for their comparative resistance and susceptibility. The assessment was based on developmental stages of pulse beetle on seeds. It was observed that the all the varieties of Vigna unguiculate were susceptible to the Callosobruchus maculatus F.

# *HYMENOLEPIS DIMINUTA* RUDOLPHI, 1819 (HYMENOLEPIDIDAE: CYCLOPHYLLIDEA) OF HOUSE RAT *RATTUS RATTUS* FROM DISTRICT HYDERABAD, SINDH, PAKISTAN.

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During present study on the assessment of parasitic diversity in House Rat *Rattus rattus* from district Hyderabad, Sindh, Pakistan, a total of 24 hosts were examined for the presence of parasites from January to August, 2019. Out of these, only 10 hosts were infected with 45 specimens of cestodes. The morphological features of these cestodes resemble with *H. diminuta* Rudolphi, 1819. Previously there is no record of *H. diminuta* Rudolphi, 1819 from Sindh province, therefore, proposed as new locality record. Present study is the part of research project no. 9412/Sindh/NRPU/R&D/HEC/2017.

# HISTOLOGICAL CHANGES IN THE INTESTINE OF MALLARD (ANAS PLATYRHYNCHOS L.) DUE TO TREMATODE NOTOCOTYLUS SINDHENSIS

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During the study period infected intestine of Mallard (*Anas platyrhynchos* L.) were processed for histological studies. Tiny pieces of the small intestine were fixed in 10% buffer formalin for 24 hrs washed several times with 70% alcohol and dehydrated through graded series of alcohol, cleared in xylene, kept in mixture of xylene and wax overnight, later transferred to pure wax for 8-10 hrs and blocks were made in L-blocks preventing air bubbles. The L-blocks were kept in a cool place for proper cooling. The blocks were trimmed and fixed on microtome and 6-8 microns thick sections were cut which after processing were stained with Haematoxylin and eosin. The histological changes in the small intestine are being reported in detail.

# PREVALENCE AND CHARACTERISTICS IDENTIFICATION OF NEMATODE; HETERAKIS PAVONIS FROM BIRD, TURDOIDES STRIATA AT SINDH – PAKISTAN

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The nematodes belong to Phylum nemathelminths, class nematode; they are mostly parasite and cylindrical elongated and unsegmented worms but cuticular may in a circular, smooth and longitudinal striation in position and complete their life cycle. The host birds were captured and dissected to find out intestinal nematode helminth worms from different localities of Sindh. For this reason, (n=54) hosts internal visceral organs were examined and (n=118) specimens were recovered from the mid-portion and terminal part of large intestine belonging to the Genus: *Heterakis* Duj., The morphological characteristic in all features comprised wider buccal capsule than deep, five well-defined lips found surrounding the mouth but female with three lips, club-shaped oesophagus wider than long narrow anteriorly contained valvular apparatus but vulva found at the anterior region of the body and whole female specimens were larger and wider than male. Whereas; the longitudinal chitinous rim and bulb were present but gubernaculum was absent. Tail pointed posterior and spicules long and unequal in size. Postanal region contained seven pairs of papillae and at the preanal region two pairs of papillae were found and the eggs were found in elliptical position. The intensity of nematode helminths was found during the month of June in the summer season when compared with months of the winter season with the consequent difference (P<0.05) among the prevalence of helminths in this bird. Such a research-based study suggested that it is a firm call for a systematic study of the helminth parasites which are continuously affecting the local birds of this region.

### DESCRIPTION AND VALIDATION OF SUB-SPECIES PLANILIZA CARINATA CARINATA AND PLANILIZA CARINATA KLUNZINGERI

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The morphometric and meristic characters and mitochondrial cytochrome c oxidase 1 (CO1) gene were used to validate the status of keeled mullet species *Planiliza carinata* and *P. klunzingeri*. Based on kimura-2 parameter,

neighbor joining tree was constructed in which *P. carinata* and *P. klunzingeri* were clustered together in a clade separated by a low bootstrap support (77%) which suggest that they are monophyletic and the morpho-anatomic variations between them might be intra-specific. In this study we confirm the subspecies status of *Planiliza carinata carinata carinata carinata klunzingeri* given by Trewavas and Ingham (1972).

#### GASTRO-INTESTINAL PARASITES IN SMALL RUMINANTS OF SWAT, DISTRICT, PAKISTAN

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This dissertation is composed after a study conducted on sheep and goats with the aim to investigate the prevalence of intestinal parasites and the species of intestinal parasites that can be found in this very livestock grazing in the selected hilly areas of Swat, for the purpose total 300 sheep and goats were selected and examined through parasitological techniques, the result of the study revealed that 187(62.33%) of the individuals studied were infested with GIT parasites and the parasites species identified were Haemonchus spp, Trichuris spp, Strongylus spp, Trichostrongylus spp, Strongyloides spp and Monezia spp. In which the highest occurring species was Haemonchus species having frequency 42 and 22.5% of the total infected population followed by Trichuris spp 31(16.6%), Strongylus spp 31(16.6%), Trichostrongylus spp 28(15%), Strongyloides spp 16 (8.6%), Monezia spp 10(5.4%),Fasciola 9(4.9%), Haemonchus+Strongylus 6(3.20%),Trichuris+Haemonchus Trichuris+Strongylus 4(2%), Trichuris+Strongyloides 4(2%) and Haemonchus+Strongyloides 2(1%). Similarly of the total 204 adults 125 were infected, 62 of 96 young individuals were infected, 93 in the total 141 goats and 94 in the total 159 sheep were infected,131 in total 214 female and 56 in total 86 male were infected. And on locality basis 41/70 ruminants from Bahrain were infested, 20/40 in chatikal, 23/39 at Gabin Jaba, 47/60 of Mandor, 27/42 Peochar and 29/49 animals from Solatanr region were found infested with parasites. For association between variables and infection chi square test was applies and, in all case, i.e gender, Age, animal species and region of the animal P>0.05 hence association of infection with any of these variables was insignificant.

### STUDY OF INTESTINAL PARASITES IN SCHOOL CHILDREN OF URBAN AREAS OF LOWER ,DIR, PAKISTAN

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Intestinal parasitic infections (IPIs) are a major cause of morbidity worldwide and have been described as an important public health problem. The present study aimed to determine the prevalence and identification of risk factors associated with IPIs among 3-12years old school age children residing in lower dir district, Pakistan from 2018- 2019. A cross-sectional school-based study was conducted using a structured pre-tested questionnaire. Anthropometric tools and stool tests were used to obtain epidemiological and disease data. The direct wet mount preparation in saline/iodine/ methods were used for stool examination. Data were analyzed using appropriate descriptive, univariate and multivariable logistic regression methods. Of the 200 children studied (mean age of8.6±3.6 years) the overall prevalence rate for intestinal parasitic infections was found to be33.3%. Children infected with single parasite accounted for 27.6% and 5.7% were detected with poly-parasitism. The study showed that helminths (21.4%) were more prevalent than protozoans (4.9%). Ascaris lumbricoides(38.5%), Hymenolepis nana (25%), Trichuris trichiura (1.3%) ,E. vermicularis(2%),),Hook worm (22.5%) cryotosporidium spp (2.15%). taxocara (1.5%), shishtosoma japanicum (0.5%) Taenia saginata (2.7%) were identified in children living in urban areas. The multiple logistic regression model indicated that age of the child, gender, family size, source of drinking water, type of milk used, house condition, feeding habit, personal hygiene and socioeconomic status were

significantly (p<0.05) associated with the IPIs. Intestinal parasites were prevalent in varying magnitude among the schoolchildren located in urban areas of lower Dir. We conclude that there is a need for mass scale campaigns to create awareness regarding health and hygiene in children and the need for development of effective poverty control programs because deworming alone is not adequate to control parasitic infections.

# INTESTINAL PARASITIC INFECTION AMONG SCHOOL CHILDREN OF SWAT, DISTRICT PAKISTAN

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Intestinal parasitic infections are distributed worldwide with ahigh prevalence especially in developing countries. Parasitic infections also causes nutritional deficiencies as parasites have a correlation with nutritional status. Infections due to intestinal parasites are very common in school aged children. The purpose of this study is to estimate the prevalence of intestinal parasitic infections and its relation with nutritional status of selected areas of primary schools of district Swat, Pkistan. The stool samples were collected from four government schools. All the samples were brought to the laboratory of Parasitology, University of Malakand, and Lower Dir Pakistan for the microscopic analysis. A total of 300 stool samples were collected. Out of the total collected samples 238 (77%) were infected with one or more than one species of parasites. In this study the single infection were 68.3% (n=205), double 8.33% (n=25), triple 3.33% (n=7) and quadrupe 0.33 %( n=1) reported. Ascaris lumbricoides 34%(n=103) followed by Trichuris trichura 14.6%(n=44), hookworm species 14.3%(n=43), Hymenolepis nana 7%(n=21), Taenia spp 4.33%(n=13), Enterobius vermicularis 3.0%(n=9), Giardia lamblia 1.33%(n=4) and Entamoeba histolytica 1%. The student from age 9-12 years were found to be most infected followed by age 5-8 years and then 13 to 15 years. It is concluded from this study that males were highly infected than females. Health education campaign should be launched for the control of such infections.

### DENGUE KNOWLEDGE, ATTITUDES AND PRACTICES AMONG MEDICAL PRACTITIONERS IN MALAKAND REGION, PAKISTSAN

#### Riaz Khan, Abdul Aziz khan, Umair Ahmad and Wali Khan\*

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Dengue fever (DF) is increasingly recognized as one of the world's major mosquito borne diseases and causes significant morbidity & mortality in most tropical & subtropical countries of the world & had become the most common arboviral diseases of human. Dengue fever is endemic in most part of Pakistan & continues to be a public health concern. Dengue vector, human knowledge & human behavior each have been reported to play an important role in the transmission of the diseases. Current study was aimed to determine the level of knowledge, attitude & practices regarding Dengue fever among the health practitioners, to study the relationship of level of knowledge & attitude with preventive practices for dengue fever. A cross sectional study was carried out in medical practitioners of the four districts of malakand division during October to November 2019. A prestructured & self-administered questionnaire was used to collect data regarding dengue fever of knowledge, attitude & practices from medical practitioners. Data was analyzed using SPSS version 20 statistical software. Most of participants have seen dengue vector (62%), the media being the most quoted source of information. Nearly 81.2% participants were aware transmission of Dengue Fever is by mosquito bite. Practices based upon preventive measures were found to be predominantly focused towards prevention of mosquito bites rather than elimination of breeding places. Although the knowledge regarding DF & mosquito control measure was quite high among the medical practitioners but this knowledge was not put into practice.

# EUPHLYCTIS CYANOPHLYCTIS SCHNEIDER, 1799 (AMPHIBIA: DICROGLOSSIDAE) IN DISTRICT LOWER DIR, PAKISTAN

#### Wali Khan<sup>1\*</sup> Sumera Naz<sup>1</sup>, Jalwa Bibi<sup>1</sup> and Noorul Hadi<sup>1,2</sup>

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Euphlyctis cyanophlyctis (the skittering frog) is one of the most widespread species in Pakistan. Present study was aimed to know the extension range of Euphlyctis cyanophlyctis in urban and rural areas of Lower Dir, the Northwestern Pakistan. A total of 33 frogs were collected, including 18 from rural and 15 from urban areas. The frogs were caught by hands covered with gloves instead of using nets. The collection was managed from August to October 2016 and from April to May 2018. Morphometric analysis, coloration as well as photographs of the frogs have been provided in detail. Skittering frogs were seen frequent in swampy areas near the water bodies. These frogs were mostly seen after sunset. Euphlyctis cyanophlyctis has also been reported from other parts in Pakistan but present record represents new to the locality.

### STUDYING THE ASSOCIATION BETWEEN BODY ROUNDNESS INDEX (BRI) AND PLASMA LIPID PROFILES

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Multiple studies have well documented the association between obesity and dyslipidemia-an irregularity in plasma lipid levels. Various body indices have been identified which serve as obesity indicators such as body mass index (BMI), waist circumference (WC) etc. Recently, a novel body index body roundness index (BRI) - has been introduced which quantifies body girth with respect to height and can also detect dyslipidemia. The present work aims to study the association between BRI and plasma lipid profiles. For the present study, the participants (n=95) were recruited from different departments of the University of the Punjab, Lahore. Participants were anthropometrically measured and interviewed, and their intravenous blood was collected. Plasma lipid levels were accordingly determined. Participants with high BRI values (>4) displayed significantly higher levels of total cholesterol (TC), triglycerides (TGs), low density lipoprotein (LDL) and non-HDL and significantly lower levels of high-density lipoprotein (HDL). The correlation analysis revealed that TC, TG, LDL and non-HDL were weak positively whereas HDL was weak negatively correlated with BRI. The association of BRI with various demographic and metabolic factors was also assessed. Significant differences were observed between BRI of four different age groups as well as BMI groups of the study population. Higher BRI values suggest irregularities in plasma lipid levels in relation to obesity. It is noteworthy that BRI detects dyslipidemia and predicts risk for cardiovascular disease (CVD). Thus, BRI can be used as an alternative to BMI as it better accounts for fat accumulation and fat distribution in the body.

# CHARACTERIZATION OF SCORPION SPECIES COLLECTED FROM BHIMBER, AZAD JAMMU AND KASHMIR

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Scorpions are among the oldest terrestrial arthropods, which are distributed worldwide, except Antarctica and some pacific islands. The study sites were selected based on different topographic condition such as foothill and mountains. Daytime collection was carried out using hand digging tools for turning stones and excavate the burrows,

as well as night samplings were done by using ultraviolet light, the collected specimens were maintained in 70% Ethanol. Morphological characterization was done on the basis of color, body shape, prosomal, mesosomal and metasomal characters. Molecular characterization was done on the basis of CO1 gene. A total sixty four specimens were collected including juveniles, young and adults. Out of which six were *Hetrometrus latimanus* and 58 were *Hottentota tamulus*. *Hetrometrus latimanus* was collected from stony land while *Hottentota tamulus* were collected under the small rocks with clay. DNA Barcode analysis showed they are closely related to already described species of respective genera.

#### INSECTICIDAL ACTIVITY OF PEGANUM HARMALA

#### Muhammad Arshad, Muhmmad Shahzad, Afshan Abbas, Razia Bashir and Sajida Naseem

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Agriculture crops are damaged by a wide variety of insect pest. This is reducing annual crop yield worldwide. Effectiveness of existing pesticide is continuously challenged by growing resistance against them. At the same time these pesticide are posing serious environmental problems and health hazards. There grew a need to establish pesticides which are eco-friendly. In this study the biopesticidal potential of ethyl acetate extract of *Peganum harmala* seeds evaluated against *Brevicoryne brassicae* (Aphididae). Extract was applied by Oral method at different concentrations (5%, 7.5% and 10%) and %age mortality was recorded at various time intervals. 100% mortality was observed at 7.5% and 10% at 18Hours and 14Hours respectively. It is concluded that the phytochemicals of *Peganum harmala* can be considered as potential candidates for developing new biopesticides.

# ANTIMICROBIAL ACTIVITY OF *PONGAMIA PINNATA*Afshan Iqbal, Muhammad Arshad, Razia Bashir, Muhmmad Shahzad, Razia Bashir and Sajida Naseem

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Antimicrobial properties of plants are well documented. Our study was design to analyze the antimicrobial potential of leaves extracts *Pongamia pinnata* Linn. *Pseudomonas aeruginosa* was collected from the University of Education Main Campus Lahore in freezed form which then regrow by using Bacterial Revival technique. Good quality of leaves of *P.pinnata* was taken from the local garden and grinded in to powdery form. Extract of the leaves was prepared in Ethanol solvent by using Ultra sonic assisted extraction method. Different concentrations of extract were used against *P. aeruginosa* to evaluate the antimicrobial activity of *P.pinnata*. It compared with commercially available antibiotic Azithromycin. Ethanol extract of *P. pinnata* showed great activity against *P.aeruginosa*. At different concentrations different zones of inhibition were examined. *P. pinnata* L has good bacteriostatic as well as bactericidal activity against hospitalized pathogen *P.aeruginosa*. Which gave maximum zone of inhibition 2.9cm at 100% concentration as compared to Azithromycin which gave 3cm zone of inhibition.

#### AVIFAUNA OF SOME SELECTED URBAN WETLANDS OF KARACHI

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The aim of present study was to show the importance of wetland in urban areas. In this study some selected urban wetlands were observed to check out the related avifauna. During the study, different waterbirds were observed in the pond of Karachi Zoo, which includes Sarus Crane, White Wagtail, Great Egret, Little Egret, Grey Leg Goose,

Mallard, crows, White throated Kingfisher, Rosy Pelican, Dalmatian Pelican, Kites and Indian Pond Heron. Similarly there was a good number of waterbirds in the lake of Safari Park, Karachi including Godwall, Ruddy shelduck, Little Egret, Cormorant, Dalmatian Pelicans, Rosy White Pelican, Grey Leg Goose, Indian Pond Heron and Night Heron, White +Throated Kingfisher and Red Wattled Lapwing. The small urban wetland of Agha Khan hospital also has certain species of birds including Grey Heron, Pond Heron, Common Sandpiper, Little Egret, Grey Leg Goose, Mallard, Cattle Egret, White Throated Kingfisher, Red Wattled Lapwing, Common Mynas and Bank Mynas. These wetlands are the man-made wetlands which are located inside the urbanized areas of Karachi. The study shows that urban wetlands are not just recreationally important, but also, they are site for residential and migratory birds, breeding ground for bird, maintains the temperature of surrounding area, also provide a site for bird watching which cannot be generally seen in urban areas. As population in urbanized areas is increasing day by day which disturbs the birds habitat, so these sites are ecofriendly for these avifauna. So, city planners and manager should increase number of such artificial wetlands to make the cities more live able, safe and environmentally friendly for both human and avifauna.

# ABSTRACTS PRESENTED AT THE FIRST VIRTUAL CONGRESS OF ZOOLOGY

(40<sup>th</sup> Pakistan Congress of Zoology – International) December 17-19, 2021

#### **SECTION - VI**

#### POSTER SESSION

# ASSESSMENT OF VARIOUS PLANTS EXTRACTSTO CONTROL THE SUCKING PEST JASSID (AMRASCA SPP) ON BRINJAL (SOLANUM MELONGENA) CROP

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In this experiment the main purpose is to screen out the bio-pesticides which is safe for human health and environment. In this experiment different biopesticde were used to control the jassid. The data were recorded after the 24 hours, 48 hours, 72 hrs and 7 days of treatment application. The RCBD (Randomized complete block design) with three replicates were used. The extracts of different biopesticde were used with the 18 % solution. The different treatments, Neem seeds, Alovera extract, Moringa leaves Extracts, Ginger Extracts, Lemon seed extracts, Eucalyptus. The minimum populations were recorded 0.98/leaf against Neem Extracts and highest populations were recorded on the Moringa leaves extracts which was 4.5/leaf. The biopesticides Eucalyptus and Ginger showed good results. The result showed that Neem extract is better with minimum post treatment infestation for field application. The means were compared by Duncan's Multiple Range Test (DMRT) at P = 0.05. IBM.

### INVESTIGATING THE DIVERSITY OF TERRESTRIALSNAIL AND THEIR HELMINTH PARASITES IN TEHSIL TIMERGARA DISTRICT DIR LOWER

#### Fakhr Ul Islam<sup>1</sup>, Kareem Ullah<sup>1</sup>, Akhtar Rasool<sup>1</sup>and Muhammad Israr<sup>2</sup>

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Snails are invertebrate belong to phylum Mollusca of the kingdom Animalia. The Molluscs are the second largest group of organisms. The snails are cosmopolitan and live in a variety of habitats such as terrestrial, freshwater and marine water. Their diversity, distribution and their effect on other organisms are not much explored in Tehsil Timergara District Dir lower. For this purpose, the present study was conducted to explore the diversity of snail and determine the helminthic parasite for which the snail act host. Snails were collected from different localities such as side of rivers, green grasses, maize fields, tomato fields, and from small plants. A total of 366 specimens were collected and preserved in 80% of ethanol in Forensic Research lab, University of Swat. For the presence of helminths parasites both water and snail alimentary canal were observed under light microscope. The snail species were identified according to the taxonomic keys. DNA was extracted from the foot muscles and alimentary canal of snail by using PCI protocol. Oxichilus alliarius, Bulimulus guadalapensis, Trochulus striolatus and Helicellia itala were identified. The highest population of Oxichilus alliaruis which is (110) and lowest population of Helicellia itala which is (74).

# INVESTIGATION OF DIVERSITY OF CHAKOORPARTRIDGE (ALECTORIS CHUKAR) IN TEHSILBABOZAI DISTRICT SWAT

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Birds are warm blooded animals and their body are covered with dense feather. These feathers act as a morphological tool which are the main character that can distinguish from the other group of animals. The skin color

of the bird is often pale pink or bluish pink and thin equal to the mammals. Chakoor are the non-migratory bird which are belong to the order Galliformes family of Phasianidae and sub family Phasianoidea. Mainly the chakoor partridge (Alectoris chukar) have 16 sub species, 7 hybrid and 24 are sub breed which are present all over the world. In Pakistan their occurrence is throughout the country, including Sindh, Baluchistan, Khyber Pakhtoonkhwa, FATA, and Gilgit Baltistan. Chakoor is a small sized gamebird which are present in a high altitude from the sea level about 1000m to 4500m on hilly rock mountains. The feather sample were identified on the basis of morphology comparing with the other mammals. On the basis of DNA barcoding is to find out the exact identity of plant and animal samples. For the first time the DNA used as a marker for the identification of the species by Paul Hebert and colleague. To investigate the dentification of chakoor partridge (Alectoris Chukar) in Tehsil Babozai of district Swat. On the basis of morphological character, we successfully identified the Alectoris chukar by microscopy observing the feather which are compare to the mammalian hair. The DNA were extracted from the 15-feather sample by using PCI method.

#### INSECTS AS ECO-FRIENDLY AND SUSTAINABLEANIMAL FEED

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Global population growth, urbanization, rising income, changes in consumption patterns, lifestyles, food preferences, rising demand for animal products, and a lack of conventional feed components all contribute to the quest for alternative protein sources for animal feed. Insect feed has minimal land and water needs and a high feed conversion efficiency into insect biomass. Insect methods of production minimize dependency on traditional feed sources while bringing valuable ingredients back into the food chain from organic waste materials, agriculture, the food industry, and other sectors. High nutritional value, feed efficiency, and reproductive capacity are all advantages of utilizing insects for livestock feed. There is a broad variety of appropriate insects, such as black army fly larvae, house fly maggots, mealworms, silkworms, locusts, grasshoppers, and crickets. Black soldier flylarvae are thought to have the highest feed potentialsince their dry weight contains up to 50% crude protein (CP), up to 35% lipids, and an amino acid profile. Insect feed is a sustainable alternative to traditional feed since insects are raised on waste streams and may provide a diverse source of revenue. Insects used in the feed may help developing nations solve socioeconomic and environmental problems, aligning with the United Nations' Sustainable Development Goals. Smallholder insect farmers have good possibilities to enhance production, improve their livelihood, and contribute to food security and a circular economy with modest initial capital inputs.

# GREENING THE DESERT: A SAFE HAVEN FORBIRD COMMUNITIES AT A DESERT ISLAND OF THE UNITED ARAB EMIRATES

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Land-use change, such as afforestation, in various parts of the world has increased to combat desertification and habitat loss. In the United Arab Emirates, extensive afforestation efforts are dedicated from decades to improve the climatic conditions and improve the carbon sequestration. Sir Bani Yas Island was declared as wildlife reserve in 1971, and an extensive afforestation project was launched to improve this desert island. With plantation of approximately two million trees and various pastures, the island started receiving more number of bird species. This study was premediated

to assess the effects of afforestation on the diversity and abundance of avifauna of the island. We compared the reports from institutional and online databases for historical diversity. From 2014-2018, the bird diversity and abundance was studied using line transect method, through stratified random sampling design, in four habitat types, i.e. planted forest, pastures, mountains and coastline. The data was subjected to Shannon-Weiner Diversity Index, Sorenson's Diversity Index, Bray-Curtis Cluster Analysis, Whittaker's beta diversity, and Chi-square Goodness of Fit Test. A total of 164 bird species were recorded from 2014-2018, belonging to 19 bird orders and 46 families. The highest number of species were recorded from the order Passeriformes with a 38.41 % of all species, followed by Charadriiformes with 29.27 %. The birds were categorised into seven feeding guilds. The highest number of species were insectivorous (35.84 %), followed by carnivorous (25.66 %), grainivorous (17.26 %), omnivorous (15.93 %), frugivorous (4.42 %), piscivorous (0.44 %), and nectivorous (0.44 %). There was a significant difference in bird diversity over the period of fou decades ( $\chi^2 = 198.73$ , df = 3, P < 0.001). Sorenson's diversity index suggested high diversity of species among various habitat types. Pastures had highest bird diversity followed by forests, coastline, and mountains according to Shannon-Weiner diversity index. The study concludes that afforestation had a significant positive impact on bird diversity and is effective intervention to improve ecological health.

### PREY PREFERENCE AND HUNTING BEHAVIOUROF ARABIAN STRIPED HYAENA (HYAENA HYAENA SULTANA) AT SIR BANI YAS ISLAND, UNITED ARAB EMIRATES

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The Arabian striped hyaena (Hyaena hyaena sultana) are nocturnal carnivores of Hyaenidae family with five subspecies based on geographical variations and are listed as near threatened (NT) under IUCN Red List of Threatened Species. Sir Bani Yas Island in UAE is home to 13 individuals of Arabian striped hyaena. Sir Bani Yas Island has 41 square kilometres protected Wildlife Park, inhabited by approximately 16000 animals belonging to 32 species of various herbivores, carnivores, reptiles, and birds. Striped hyaena is reported mainly as scavenger but occasionally hunt prey up to the size of a barbary sheep (Ammotragus lervia). In the current study, we aimed to record the prey preference of Arabian striped hyaena when abundant prey population and variety is available in space limited wildlife reserve. The data on prey species, sex, age, degree of consumption, date and time of kill, and kill location was collected between 2014 and 2016, through direct and indirect observations. The most abundant prey species was Arabian sand gazelle (Gazelle subgutturosa marica) comprising about 79 % of all predated species, followed by 6 % each for both blackbuck (Antilope cervicapra) and mountain gazelle (Gazella gazella cora), axis deer (Axis axis) 5 %, and 2 % each for barbary sheep and Arabian tahr (Arabitragus jayakari). Arabian striped hyaena predated more males (55 %) compared to females (45%). Most of the kills were recorded during the early morning (56 %) followed by 21 % at night, 13 % during the afternoon, and 10 % in the evening. Moreover, adultprey was preferred by Arabian striped hyaena (89 %) as compared to sub-adult (6 %) and juveniles (5 %). The preliminary results of the current study suggest that Arabian striped hyaena prefer to hunt if there are abundance and diversity of prey species around them.

# PHYSIOLOGICAL RESPONSES OF CAFFEINE ONSMOOTH MUSCLE: A STUDY ON REPTILE UROMASTIX *HARDWICKII*

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Caffeine, a metabolic stimulant provides useful information on sarcoplasmic reticulum's properties in a muscle tissue. Intestinal segment ileum, from different classes of vertebrates has been studied for the functional differences using Caffeine as a pharmacological tool. This study was designed to determine the effect of caffeine concentration on the mechanical response of smooth muscle (ileum) from Uromastix and Rabbit. Present investigation is a continuation of previous studies carried out by the author. Temperature controlled bathing using isolated organ bath assembly with isometric transducer was employed to conduct the experiments. Mechanical records in response to varying concentrations (0.5mM – 10mM) of caffeine were obtained digitally using Power Lab, from the of ileum Uromastix and Rabbit. A gradual decrease in basal tone of non rhythmic ileum from Uromastix was observed using low to high caffeine concentration. While spontaneously rhythmic active ileum of Rabbit, showed a gradual decrease in its active tension when applied with high caffeine concentration and vice versa. In the light of above investigation, it is concluded that caffeine may not necessarily induce tension development in reptilian model. Further, in mammalian tissues the concentration of caffeine does play an important role in its mechanical response.

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